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ORIGINAL ARTICLES

NEPHROTIC SYNDROME

Effect of Adrenocorticotrophic Hormone

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DURING the past year preparations capable of stimulating adrenal cortical activity or of simulating its effects have been investigated with regard to their action in a wide variety of diseases. Although many investigators have been using these materials in the nephrotic syndrome, published reports are few.[1-3] This report will be concerned primarily with clinical and metabolic observations on 14 patients with the nephrotic syndrome while under and following treatment with adrenocorticotrophic hormone (ACTH—Armour*).

The reasons for attempting to treat this disorder with ACTH were highly speculative. Among them was the thought that the effect of rubeola and some other infections in inducing diuresis might be the result of adrenal stimulation by the "stress mechanism" of Selye.[4] A second consideration was that nonnephrotic patients receiving ACTH had frequently been observed[5] to retain salt and water, and subsequently to have a diuresis while still receiving treatment or following withdrawal of medication. The third and more general consideration was that ACTH is effective in many chronic diseases capable of spontaneous remission; the nephrotic syndrome is an example of such a situation.

CLINICAL MATERIAL

All the 14 patients were under 16 yr. of age and most of them were in the usual age group of nephrotic patients, 2 to 5 yr. All 14 showed the classical findings of the nephrotic syndrome: anasarca, hypoproteinemia, massive proteinuria and hypercholesterinemia. No attempt was made to

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TABLE 1

GROUP A(1)—COMPLETE CLEARING OF EDEMA

Patient	Age at onset of nephrosis	Duration of disease before 1st ACTH	No. of previous diureses	Causes of previous diureses	Time since last edema-free	Signs of "Nephritis"*		
						Hematuria	N retention	Hypertension
R.C. 3 7/12 yr.	19 mo.	24 mo.	1	Spontaneous	6 mo.	++	++	+
L.D. 3 yr.	24 mo.	12 mo.	1	Spontaneous	11 mo.	+	+	+
R.dG. 2 5/12 yr.	21 mo.	8 mo.	2	1. Spontaneous 2. Rubeola	3 mo.	+	+	++ (at onset)
L.M. 1 8/12 yr.	20 mo.	2 wk.	0	—	2 wk.	0	0	0
R.F. 2 3/12 yr.	24 mo.	3 mo.	1	Rubeola	1 mo.	+++	++	+
GROUP A(2)—SIGNIFICANT BUT INCOMPLETE CLEARING OF EDEMA								
S.H. 16 2/12 yr.	15 10/12 yr.	4 mo.	0	—	4 mo.	+	+++	++
R.R. 3 7/12 yr.	15 mo.	28 mo.	1	Na and K feeding	Never edema-free. 12 mo. since diuresis	0	0	0
E.G. 4 yr.	36 mo.	12 mo.	0	—	12 mo.	+++	?	+ (?)
W.D. 2 11/12 yr.	28 mo.	7 mo.	1	Yellow fever vaccine	4 mo.	+	0	+
GROUP B—NO SIGNIFICANT CHANGE IN EDEMA								
P.F. 5 8/12 yr.	41 mo.	27 mo.	2	1. Spontaneous 2. Rubeola	17 mo.	++	0	+
C.M. 4 1/12 yr.	31 mo.	18 mo.	1	Spontaneous	5 mo.	++	+	++
M.R. 10 4/12 yr.	9 10/12 yr.	6 mo.	2	1. Spontaneous 2. NH_4Cl and plasma	4 mo.	+	++	±
B.L. 4 4/12 yr.	37 mo.	15 mo.	5	1, 2, 3, 4. Spontaneous 5. Rubeola	6 mo.	+	+	+
F.deM. 2 4/12 yr.	15 mo.	13 mo.	2	1. Spontaneous 2. Rubeola	4 wk.	++	+	+

**Explanation of Symbols Under "Signs of Nephritis"*

Hematuria
+ implies: Occasional—10 RBC/hpf in sedimented urine
++ implies: 11–30 RBC/hpf in sedimented urine
+++ implies: 31–60 RBC/hpf in sedimented urine
++++ implies: >60 RBC/hpf in sedimented urine

N Retention
+ implies: Blood NPN 40 to 55 mg./100 cc. on 2 or more occasions
++ implies: Blood NPN 56–70 mg./100 cc. on 2 or more occasions
+++ implies: Blood NPN 71–100 mg./100 cc. on 2 or more occasions
++++ implies: Blood NPN >100 mg./100 cc. on 2 or more occasions

Hypertension
+ implies: diastolic arterial pressure 85 to 95 mm.Hg on several occasions
++ implies: diastolic arterial pressure 96–105 mm.Hg on several occasions
+++ implies: diastolic arterial pressure 106–115 mm.Hg on several occasions
++++ implies: diastolic arterial pressure >115 mm.Hg on several occasions

N.B. Findings of 1+ in the author's experience are of little prognostic significance.

draw a sharp distinction between "pure nephrosis" and "the nephrotic phase of chronic glomerulonephritis"; however, none of the patients showed evidence of severe renal damage and insufficiency at the time ACTH was given. Further details are best seen in table 1.

For purposes of discussion the patients have been grouped according to their response to treatment: Group A_1, those who completely lost their edema in close time relationship to treatment; Group A_2, those who had significant diuresis but who failed to become edema-free; Group B, those who had no significant benefit.

Plan of Study

All patients were kept on a fairly constant low sodium and chloride intake, no salt being added in the preparation of food for consumption. Where complete urine collections were possible the diets were weighed and Na, K and Cl intakes calculated from standard tables.[6,7] For the small children the sodium intake was of the order of 10 to 14 mEq./day, chloride 12 to 18 mEq./day and potassium 40 to 60 mEq./day. When possible, total daily urinary output of Na, Cl, K and protein was determined; when this could not be done, concentrations of these substances in the specimens collected were measured. Blood plasma was examined before, during and after treatment for the concentration of protein and cholesterol. It was examined twice a week for the levels of Na, K, Cl and CO_2. Hematocrits also were determined twice a week.

The hormone was administered intramuscularly every 6 hr. for 4 to 6 day "courses." For the most part the individual dose was 15 mg. (60 mg./day), the larger children receiving a somewhat higher dose. Individual daily total doses used are shown in chart 1.

Technical Methods

Sodium and K determinations were made on an internal standard flame photometer[8] constructed in this laboratory, chloride by the Schales method,[9] cholesterol by the Schoenheimer-Sperry method[10] and urinary protein by the Shevky-Stafford technic.[11] Serum protein partition was done by the Howe method.[12] Daily absolute eosinophil counts were done by the method of Henneman et al.[13]

Clinical Observations

Of the 14 patients treated five had copious diuresis which resulted in complete delivery of their edema in close time relationship to the administration of ACTH; four had significant diuresis which stopped before they were edema-free; five showed no noteworthy effect.

Several of the patients were given two courses of the drug. Two (R.C. and L.M.) were in Group A_1, having lost all edema after the first course; symptomatic relapse occurred after several weeks and a second course resulted in a response similar to the first. A third (R.F.), who had an initial good response, began to reaccumulate edema immediately and a second course at this time had no observable effect. Two patients (S.H. and R.R.), who partially lost their edema with the first course, were treated again when the diuresis had stopped. The response on these occasions was also incomplete. Two others (P.F. and C.M.), who showed no response on the first occasion, were immediately given a second course, still with no response. Chart 1 shows the duration of the courses and the time intervals between courses.

No untoward effects were observed which could be attributed to the production of a hyperadrenocortical state, except in one patient, B.L., who was treated for almost eight days. Near the end of the period of therapy generalized tremors developed and simultaneously the plasma concentration of potassium was found to be 2.8 mEq./l. Hypokalemia is a known feature in hyperadrenocorticism.

Follow-up on these patients varied from 7 to 15 months. Of those in Group A_1, R.C. and L.D. have remained essentially edema-free for 12 and 11 months. L.D. had rubeola after she had lost her swelling. Both have been on sodium lactate by mouth, approximately 100 mEq./day as suggested by Fox and McCune,[14] and both still show marked proteinuria. L.M. has had repeated recurrence of edema about every three months

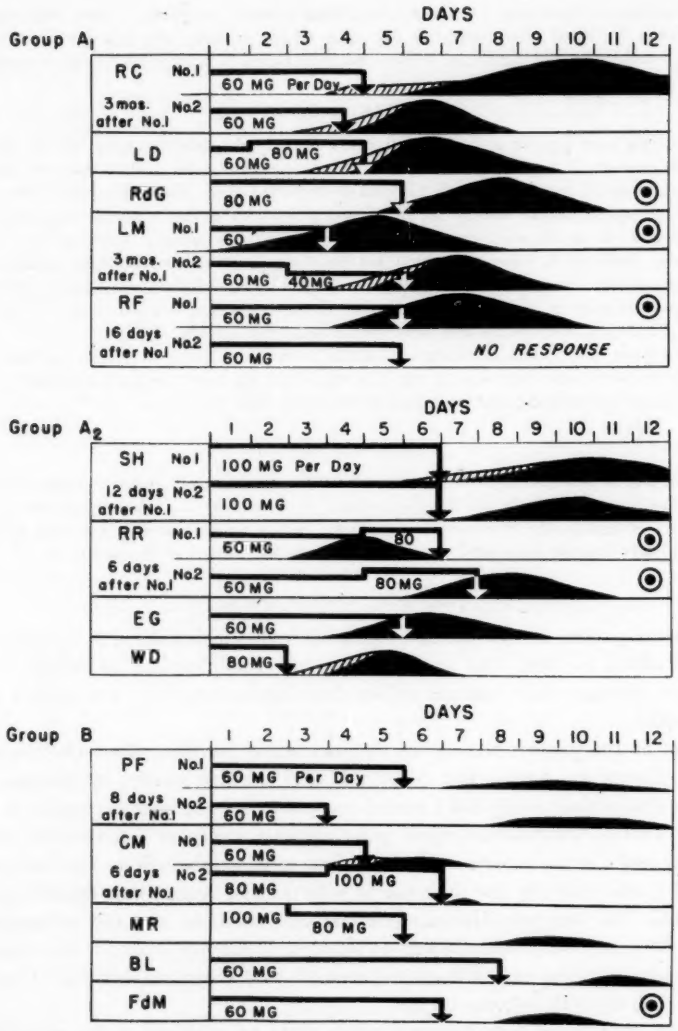

CHART 1. Diagram to show ACTH dosage and time relationship between therapy and response. Hatched areas represent onset of water diuresis, black areas sodium plus water diuresis. Height of areas is not quantitative but indicates approximate intensity of diuresis. Daily dose of ACTH shown under arrow. Bulls-eye symbols indicate urine collection incomplete so that onset of water diuresis could not be determined.

and has currently responded four times to ACTH. Sodium lactate was given to her following one such diuresis but she reaccumulated fluid so this was discontinued. R.dG. and R.F. both had rapid recurrences and failed to respond to later courses.

Three of the patients in Group A_2 subsequently lost all edema (R.R., W.D. and E.G.)

while taking sodium lactate and have been symptom-free for 8, 9 and 10 months. Of these W.D. had intercurrent rubeola while edema-free. R.R. has recently had urine free of protein and may be going into permanent remission. S.H. continues to have edema but now has responded in some degree to four separate periods of treatment.

Those in Group B have had varied courses. P.F. had a spontaneous remission three months after treatment which lasted six months. After edema recurred he failed to respond to still another trial of ACTH. C.M. continued in his edematous state, but about three months later another period of treatment initiated a transient diuresis. Following this he was given sodium lactate, lost the rest of his edema, but relapsed after about three months. M.R. has run a course characterized by progressive renal impairment and currently, seven months later, is in frank uremia. B.L. has continued without remission and failed to respond to another trial on ACTH; he is beginning to show evidence of renal failure. F.dM. was treated with cortisone following the failure on ACTH and has for the past seven months been edema-free though proteinuria persists.

From these observations of the post-treatment course of the disease few generalizations are justified. It has proved impossible to predict which patients would respond to treatment. It is apparent that in occasional patients the edema can be controlled at least temporarily by repeated administration of the drug. Whether such management will alter the eventual prognosis for this condition can only be determined by a much longer period of study. Another question still unanswered is whether longer periods of treatment may prove more effective. In this series prolonged administration was avoided because of the danger of secondary undesirable effects of hyperadrenocorticism.

Biochemical Observations

When diuresis occurred in close time relationship to ACTH administration, the outpouring of water and salt was comparable to that seen whenever diuresis occurs. An illustrative case of spontaneous diuresis appearing in a 3 year old boy with the nephrotic syndrome is seen in chart 2 and table 2. He was admitted for study and possibly ACTH treatment, but began to lose edema before any therapy was given. Charts 3 and 4 and tables 3 and 4 demonstrate what happened in the case of R.C. with two courses of ACTH. It will be seen that as far as water and electrolyte output are concerned the diuresis was of the same type as that seen in the spontaneous instance shown above. On the second occasion (chart 4), as the output of sodium began to diminish, the patient was given orally extra sodium in the form of the acetate, the dose being increased to about 100 mEq./day. At this time, despite the fact that there was no significant change in his serum protein level (table 4), edema did not accumulate and the increased sodium intake was promptly reflected by a rise in the urinary sodium. Extra sodium was given to all patients who showed good or partial response except R.F. and S.H. and in all of these the ability to excrete sodium was similar to that exhibited by Patient R.C. This implies that the kidney in these cases was at least temporarily capable of handling increased sodium loads in a fashion which appears normal when compared to the data presented by Leaf et al.[15] The material shown in these charts is very similar to the findings in all the patients who had any significant response to treatment.

Chart 5 and table 5 are presented to demonstrate the fact that even in the absence of a clinically significant response there is, nevertheless, a slight sodium diuresis. Better to show this fact, the ordinate for sodium in this chart has been increased by a factor of 5.

This is representative of the phenomena observed in all except one (R.F., second course) of those patients who showed no clinically observable effect.

The time relationship between the administration of ACTH and the beginning of diuresis is depicted in chart 1. In charts 3 and 4 a very slight increase in urine volume

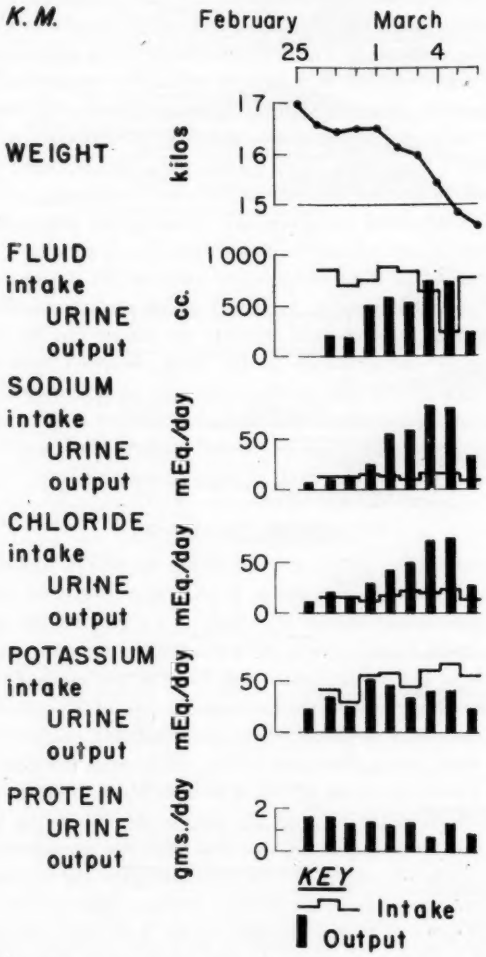

CHART 2. Chart showing spontaneous diuresis in 3 yr. old child with nephrotic syndrome. Weight, intake and urinary output shown here. Blood findings in table 2.

two days before the increase in urinary sodium is evident. In a single case this would not be significant, but it happened in so many cases that it was the author's impression that it was a characteristic feature. Accordingly, in chart 1 water diuresis has been plotted independently of sodium diuresis, so far as the data permitted. Only in the case of S.H., second course, did water diuresis begin after sodium diuresis. From this diagram it is

seen that in every instance where significant diuresis occurred it began while the drug was still being administered. In those patients in whom complete clearing of edema resulted, diuresis seemed to begin earlier than in those with only partial response. In those with no significant response, the slight measurable sodium diuresis was greatly delayed. In general, when there was a satisfactory result, it began 4 to 6 days after starting the drug.

In six instances the measurement of urine volume and of food intake was sufficiently complete to allow calculation of the negative balance of sodium and chloride during diuresis, at least as represented by that portion excreted in the urine. In these cases the ratio of sodium loss to chloride loss measured in milliequivalents averaged 1.13, with a

TABLE 2

BLOOD FINDINGS ON PATIENT K.M.
(Other data shown in chart 2)

	February				March					
	25	26	27	28	1	2	3	4	5	6
PLASMA										
mEq./l Na			136.9			133.8				138
mEq./l K			5.15			5.0				5.6
mEq./l Cl			104			105				105
mEq./l CO_2			23.3			24.6				23.7
SERUM										
gm./100 cc. Alb.			1.89							
gm./100 cc. Glob.			2.26							
mg./100 cc. NPN			29							
HEMATOCRIT %			42.2			42.5				41.1
Circ. Eos./cmm.			34	56	91	50	122			

range from 0.99 to 1.34. The excessive loss of chloride, as reported by Fox and McCune[14] in one instance of a patient undergoing a comparable diuresis, was not observed.

The amount of urinary protein excreted per day showed consistently some slight diminution during the period of diuresis, but the change was small. This is in contrast to the striking drop in proteinuria reported by Janeway et al.[16] in the diuresis which follows rubeola. The daily potassium excretion did not vary consistently during diuresis.

The level of sodium in the plasma of the 14 patients before treatment averaged 133 mEq./l. with a range from 126 to 140. Change in this level during treatment was usually in the direction of increase, but was of small degree. The range of change was from −5.9 mEq./l to +16.2, with an average of +3, which seems not to be significant. Two of the cases in which there was a fall in plasma sodium concentration following ACTH treatment were in the group which showed diuresis; three others with no change or a decrease were in the group in which no response to treatment was seen.

The level of plasma bicarbonate was usually low to start with, in the order of 20-23 mM/l., and tended to rise a little with treatment. Plasma chloride concentrations regularly showed a slight fall. Those of potassium did not show any consistent change.

In 13 instances out of the 21 courses of treatment given, the total serum cholesterol concentration was measured before and a few days after the completion of the course. In 10 of these there was a significant fall amounting to 20 to 30% of the initial value—in one instance 45%. This was not strictly related to the clinical response, two patients failing to exhibit diuresis in spite of cholesterol decreases of 21 and 39%.

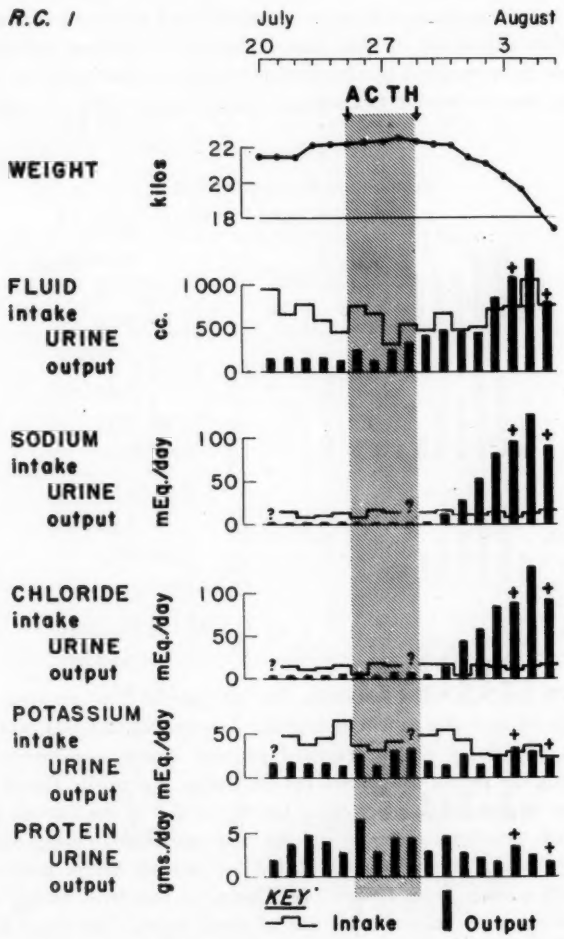

CHART 3. Chart showing diuresis in R. C. induced by ACTH, first course. Other data in table 3.

Serum protein concentration and the ratio of albumin to globulin occasionally showed some change toward normal, but of small degree. The failure of such change to occur was not inconsistent with a favorable response, as can be seen in tables 3 and 4.

Hematocrit readings were made before and after treatment in all cases, but as they were done only twice a week it is entirely possible that significant changes were missed.

In 7 of the 12 instances in which diuresis resulted, there was a concomitant drop in hematocrit ranging from 5 to 32% of the initial figure. In 3 of the remaining 5 in this group there was a rise of 10 to 20%, and in the others no change. In the cases giving no clinical response there was a fall of 5 to 16% in 6 out of 9 instances, and no change in the others.

Absolute eosinophil counts were done in all cases as a measure of the effect of the ACTH. In all instances there was a striking drop almost to zero. In some instances it rose to levels higher than before treatment upon withdrawal of the hormone.

Discussion

The facts here presented show too much variation to permit any interpretation of the action of ACTH in the nephrotic syndrome. The author has seen no features common to

TABLE 3

Blood Findings on Patient R.C., First Course of ACTH
(Other data in chart 3)

		July											August				
		20	21	22	23	24	25	26	27	28	29	30	31	1	2	3	4	5
PLASMA																		
mEq./l.	Na	134		140			142		136		137			143		137		139
	K	4.0		4.6			4.7		4.8		4.4			4.6		4.6		4.7
	Cl	109		112			108		107		109			111		110		113
	CO_2	20.5		20.9			20.5		20		18.6			23.3		23.0		23.6
SERUM																		
gm./100 cc.	Alb.	1.3								1.1								1.4
	Glob.	2.2								2.4								2.6
mg./100 cc.	Chol.		552							440								345
	NPN	33		39			40			42						31		22
HEMATOCRIT %		39.8		39					40.?					39.9		39		37.1
Circ. Eos./cmm.			166	72	59		75	0	0	0	0	28		153	85	63	28	53
							↑ ACTH ↑											

the members of any group of patients which would make it possible to predict those most likely to have a favorable response to treatment. Specifically, frequency of manifestations such as hematuria, azotemia and hypertension, commonly thought to imply the existence of "nephritis" was about the same in those in whom the outcome was satisfactory and in those in whom it was not. Duration of the disease prior to treatment was not significantly different in any group. Age of the patient at the time of onset of the syndrome may possibly prove to have some bearing, in that all those in the group which responded most satisfactorily began their illness at a relatively young age (19 to 24 months) whereas those with partial or no response were all older at onset with the exception of Patient F.deM.; she, it must be noted, did apparently respond later to cortisone. The group is too small, however, to draw any definite conclusions in this respect.

None of the data suggest the mechanism by which the hormone induces diuresis. The hypothesis that there might be a period of adrenocortical hypofunction upon cessation of the stimulating effect of exogenous ACTH, and that this might be the reason for the

outpouring of salt and water has been advanced. This would seem to be denied by the fact that diuresis occurred frequently while the patient was still receiving the material. In one patient, L.M., it began almost as soon as the drug was started so that in this case at least adrenocortical hyperfunction certainly did nothing to interfere with diuresis. The data do show that at least temporarily following treatment, in the successful cases, the kidney is normal with respect to its ability to excrete sodium.

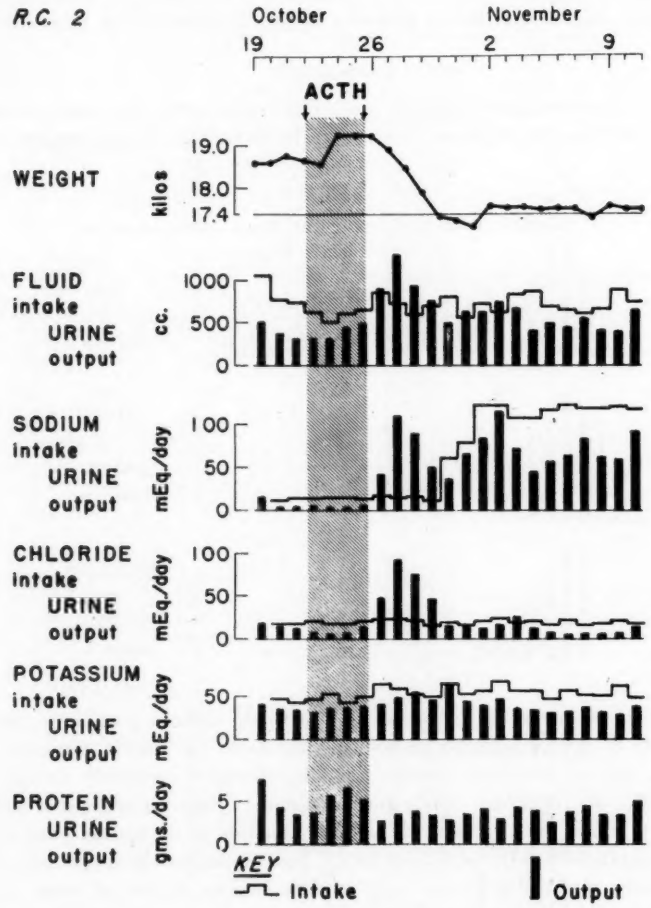

CHART 4. Chart showing diuresis in R. C. induced by ACTH, second course. Other data in table 4.

It is obvious that no evaluation of the effect of ACTH on the underlying disease process in the nephrotic syndrome can be made. Longer follow-up of patients treated repeatedly will be required for this purpose. Nevertheless, it appears that diuresis is produced with far greater frequency by the administration of ACTH than with any known agent except rubeola. It must be emphasized that nothing suggesting cure of the underlying disease has so far been seen. But even if not curative, ACTH permits the

TABLE 4

BLOOD FINDINGS ON PATIENT R.C., SECOND COURSE OF ACTH, 3 MONTHS AFTER FIRST
(Other data shown in chart 4)

		October													November									
		19	20	21	22	23	24	25	26	27	28	29	30	31	1	2	3	4	5	6	7	8	9
PLASMA																							
mEq./l.	Na	140							141		141.5			136.5		140.4		137.8			141.6		140
	K	6.5							5.87		7.17			4.02		4.25		4.45			4.81		4.91
	Cl	110							109		102			102		108		107			107		102
	CO_2	22.7							25.3		27.1			27		29		25.9			23.2		25.9
SERUM																							
gm./100 cc.	Alb.	0.86																			1.5		
	Glob.	2.97																			2.94		
mg./100 cc.	Chol.	566																			878		
	NPN	40							41												38		
HEMATOCRIT %		48.6							44.3		43.3			43.3							41.6		42
Circ. Eos./cmm.			325	272	237	87	18	33	327	312	317			460		98	135	247			197		203

↑— ACTH —↑

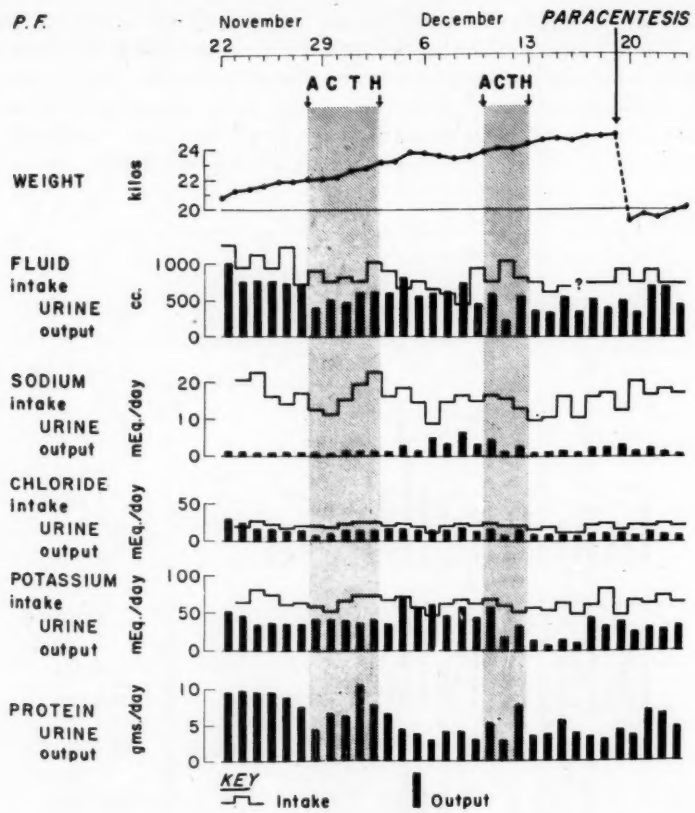

CHART 5. Chart showing electrolyte output in P. F. associated with ACTH. No significant diuresis. Ordinate for sodium increased by factor of 5 as compared with previous charts. Other data in table 5.

induction of diuresis with fair regularity under controlled conditions and thus should prove a useful tool in the investigation of the nature of the edema and the initiation of diuresis.

SUMMARY AND CONCLUSIONS

The effects of short term ACTH treatment (4 to 6 days) in 14 patients with the nephrotic syndrome are presented. Five patients given eight courses showed complete clearing of edema in seven instances, one failing to show any response to a second course. Four patients given six courses showed significant but incomplete clearing of edema each time. Five patients showed no clinically significant response to seven courses.

The type of diuresis with regard to salt and water excretion was similar to that seen with spontaneous diuresis. In all but one instance the patients who failed to have a clinically significant response did, nevertheless, show a slight sodium diuresis after withdrawal of the drug. There were no consistent changes in the significant chemical findings in the blood, except that in most cases, regardless of clinical results, a drop in the level of total cholesterol was observed after treatment. Several patients who received sodium

TABLE 5

BLOOD FINDINGS ON PATIENT P.F.
(Other data shown in chart 5)

	November									December																						
	22	23	24	25	26	27	28	29	30	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
PLASMA																																	
mEq./l. Na		123			126		125					127		133		133			137		138		138			136		137		136			134
mEq./l. K		4.5			4.2		4.5					3.9		3.5		3.2			3.1		3.2		4.1			3.0		3.4		3.5			3.7
mEq./l. Cl		96			100		101					98		105		101			105		102		102			104		102		104			100
mEq./l. CO_2		22.2			22.3		24.1					25.0		22.7		23.5			27.8		23.9		26.6			26.5		26.4		26.2			26.3
SERUM																																	
gm./100 cc. Alb.		1.1										1.4																					
gm./100 cc. Glob.		2.4										2.5																					
mg./100 cc. Chol.		820										820																					
mg./100 cc. NPN		24										38															32						34
HEMATOCRIT %		31.5			31.3		31.3					38.4		34.7		34.5			32.4		37.1		37.3			37.9				37.5			39.6
Circ. Eos./cmm.		62			112	109	73	50	16	6	31	57	73	70	70	78			100	31	7	8	210	431	390	291	261	134	153	162	131	169	
							↑— ACTH —↑												↑— ACTH —↑														

lactate by mouth following diuresis did not accumulate edema and had a pronounced increase in the urinary sodium. This suggests that at least temporarily their kidneys were capable of dealing with sodium in normal fashion.

The period of post-treatment studies on these patients varies from 7 to 15 months. Some of the patients have remained symptom-free, others have relapsed several times but responded to subsequent periods of treatment, still others after early successes have failed to respond to later ACTH administration. Only one of the group has shown clearing of the urine suggesting a permanent remission. Thus the use of this hormone at best offers only a means of controlling edema of the nephrotic syndrome. Whether its repeated use may alter the over-all prognosis can only be determined after a much longer period of observation. It should, however, prove useful in the study of the mechanism of diuresis.

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Spanish Abstract

Efectos de la Hormona Adrenocorticotropica (ACTH) en el Sindróme Nefrótico

El autor describe los efectos obtenidos con un tratamiento corto de ACTH, (4 a 5 días) en catorce pacientes con sindróme nefrótico. En cinco pacientes que recibieron ocho cursos de ACTH, en siete ocasiones se produjo una diuresis con desaparición completa del edema y un paciente no respondió a

la administración de un segundo curso de la droga. En cuatro pacientes que recibieron seís cursos de ACTH, en todos se observó una desaparición incompleta del edema de grado significativo. En cinco pacientes que recibieron siete cursos de ACTH, no se demostraron cambios clinicos importantes.

El tipo de diuresis observado en relación a la excreción de agua y sales, fué similar a la diuresis espontánea. Una pequeña diuresis de sodio fué observada a la terminación del tratamiento en aquellos pacientes que no demostraron cambios clinicos importantes, a excepción de un caso. No se notaron cambios consistentes de la química sanguinea a excepción de una caida de los niveles del colesterol ocurrida después del tratamiento, independientemente de los resultados clinicos. En varios pacientes que siguiente a la diuresis recibieron lactato de sodio por la via oral, no se demostró acumulación de edema y si un aumento marcado de la excreción urinaria del sodio. Esto sugiere que los riñones son capaces de manejar el sodio en una manera normal, al menos temporalmente.

El periodo post-tratamiento de este grupo de pacientes varió entre siete y quince meses. Algunos de ellos han permanecido libre de síntomas, otros han experimentado varias recurrencias de la enfermedad, la cual respondió a tratamientos ulteriores, y finalmente algunos pacientes que habiendo respondido favorablemente al tratamiento inicial, fueron refractarios a tratamientos ulteriores.

Del grupo total de pacientes estudiados, solamente uno ha permanecido con la orina libre de albúmina, lo que sugiere la ocurrencia de una remision permanente. El autor estima que la utilidad de esta droga consiste en el manejo y control del edema del sindrome nefrotico. En cuanto a si el uso repetido de esta medicación pueda alterar favorablemente el pronóstico-de la enfermedad, solo podria determinarse después de un periodo grande de observación. Sin embargo, su empleo debe ser de utilidad practica en el estudio del mecanismo de la diuresis.

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RESPONSE OF THE SERUM HYALURONIDASE INHIBITOR AND MUCOPROTEINS TO ADRENOCORTICOTROPIC HORMONE IN RHEUMATIC STATES

Mucolytic Enzyme Systems XV

By FORREST H. ADAMS, M.D., VINCENT C. KELLEY, M.D., PAUL F. DWAN, M.D., AND DAVID GLICK, PH.D.
Minneapolis

THE role of hyaluronidase as a "spreading factor" in infectious diseases has been reviewed repeatedly.[1-3] The proteins of hyaluronidase can act as an antigen, and the specific antibodies which can be elicited inhibit the activity of the antigenic enzyme but not the enzyme from other sources. In addition to this specific antibody inhibitor, the blood serum of animals and man contains a nonspecific inhibitor which can affect hyaluronidase from any source. Previous papers in this series have dealt with the increase in concentration of the nonspecific inhibitor in serum in a wide variety of infectious diseases,[4-7] including rheumatic conditions, cancer,[8-10] liver[11] and kidney[12] disease. An integration of these investigations as well as others is given in a recent review.[13]

It has been shown by Winzler and coworkers[14, 15] that the mucoprotein concentration in the serum is elevated in pneumonia and cancer. Kelley and Good[16] also observed an increase in rheumatic disease. Distinction between the mucoprotein and the hyaluronidase inhibitor of the serum was brought out by Glick et al.[17]

The role of the adrenal cortex as a possible mediator and regulator of the blood levels of the hyaluronidase inhibitor and the mucoproteins was indicated in a preliminary report;[18] and from other experiments,[19] it was demonstrated that administration of adrenocorticotropic hormone (ACTH*) to normal animals and to hypoglycemic children resulted in an elevation in the serum concentrations of both these factors. In view of the demonstration of the effectiveness of adrenocorticotropic hormone therapy in the same diseases in which these serum values are elevated,[20] it seemed of importance to further elucidate any relationship that might exist. For this reason, the present study of the response of the nonspecific hyaluronidase inhibitor and the mucoproteins to the administration of ACTH in patients acutely ill with rheumatic fever and rheumatoid arthritis was undertaken.

MATERIALS AND METHODS OF STUDY

Thirteen patients with acute rheumatic fever and 2 with juvenile rheumatoid arthritis were the subjects of this investigation.† One of the patients with acute rheumatic fever also had diabetes

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* ACTH used in these studies was obtained from Armour Laboratories, Chicago.

† Since the presentation of this paper an additional 15 children with rheumatic states have received either ACTH or cortisone and have been studied in a similar manner. In general the results have been the same as those reported in this communication.

mellitus which had been present previous to the onset of his rheumatic fever. The ages of the patients ranged from 4 to 17 yr. Following a control period of 1 to 5 days, ACTH was administered intramuscularly in doses ranging from 40 to 100 mg./day. The average duration of the treatment was approximately 4 wk. Fasting blood specimens were obtained every day or every other day for the following determinations: hemoglobin, eosinophil count, sedimentation rate, nonspecific hyaluronidase inhibitor and mucoprotein-tyrosine. Sera were likewise saved at critical periods of the study for electrophoretic and total protein analysis. The following additional tests or studies were made of secondary interest: daily electrocardiograms, electroencephalograms, determinations of heart size and psychiatric evaluation.

The nonspecific hyaluronidase inhibitor was determined by the viscosometric method described previously,[6] with later modifications.[21, 22] The values were recorded as per cent inhibition. The mucoprotein tyrosine was determined by the method of Winzler et al.[31] using the Folin-Ciocalteau reagent. Eosinophil counts were done according to the technic described by Randolph.[23] Counts were made on specimens removed both before and after the administration of ACTH; however, the values recorded on the graphs to follow were obtained from the fasting morning specimen (in each case before the drug was given for that day).

Results

In general the sedimentation rate and nonspecific hyaluronidase inhibitor level dropped rapidly during the administration of ACTH usually to normal values within 10 to 15 days. This seemed to follow quite closely the subjective improvement in the patient's condition. The ACTH effected not only a quick drop in the hyaluronidase inhibitor level but in all instances the level became subnormal. Such subnormal values had previously been observed in patients with inactive rheumatic fever.[7]

The serum mucoproteins, however, were much slower in their return toward normal levels. This usually took 3 to 4 months. This is in contrast to the observations in patients with acute rheumatic fever who had not received ACTH; such an example is shown in chart 1. Under such conditions, the serum levels of the mucoproteins and the hyaluronidase inhibitor paralleled each other quite closely.

In instances in which the ACTH was withdrawn or the dosage was cut too low before a substantial drop of the mucoprotein level had taken place, the patient had a recurrence of the disease. This occurred even though the dosage was not changed until the sedimentation rate and the hyaluronidase inhibitor level were normal. In general, the fasting eosinophil count increased during ACTH therapy at the same time the inhibitor concentration and the sedimentation rate fell to normal.

ACTH administered to the patient with combined diabetes mellitus and rheumatic fever effected an increase in the insulin requirements, approximately double that of the pre-ACTH period. As the ACTH was gradually withdrawn, the insulin requirement returned to the previous amount.

Clinically the ACTH relieved the patients of their acute symptoms as has been observed by others. It was felt that the duration of the disease was probably shortened and that the cardiac disability was kept to a minimum and was prevented from progressing. However, it should be noted that many patients with acute rheumatic fever have a similar course without therapy. Most of the heart murmurs disappeared during the therapy or were so minimal that when the patient was checked again in the outpatient clinic, none could be ascertained except for those who had had a cardiac lesion previous to the episode of rheumatic fever for which the patient was being treated.

No serious complications from the administration of ACTH were encountered in any of the patients. Mild acne over the forehead was observed in about half the patients.

All the children tended to retain fluid in the tissues to a mild degree, but not sufficient to produce pitting edema. No adverse psychiatric or cardiovascular effects were encountered.

Illustrative cases indicating the type of response and the general problems involved follow:

Patient E. R.: This was a 13 yr. old female who had been bedridden 4 yr. with rheumatoid arthritis. Photographs before and after 1 mo. of ACTH therapy can be seen in figure 1. This

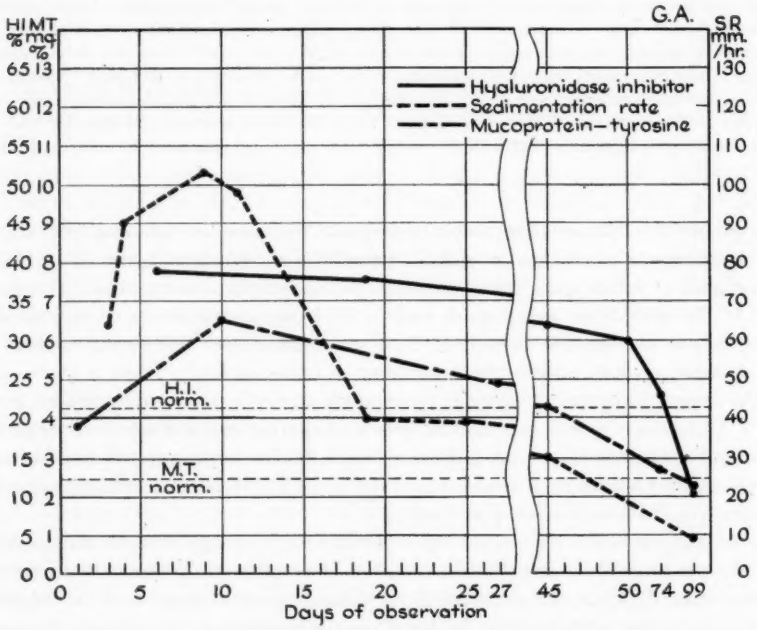

CHART 1. Patient G. A., 11 yr. old boy with acute rheumatic fever treated symptomatically with salicylates. In this and following graphs, nonspecific hyaluronidase inhibitor (H.I.) is expressed as per cent inhibition, mucoprotein tyrosine (M.T.) as mg./100 cc. and erythrocyte sedimentation rate (S.R.) in mm./hr.

patient experienced a remarkable relief of her arthralgia within 6 hr. after the institution of ACTH 50 mg./day. Previous to ACTH therapy her daily caloric intake of food had been between 600 to 700 calories, so low that she had been seen by the psychiatrists as possibly having anorexia nervosa. On ACTH therapy her caloric intake rose to approximately 3,000 cal./day; this occurred concomitantly with the general improvement in her psyche. The sharp drop to normal of the sedimentation rate and the hyaluronidase inhibitor within 5 days can be seen in chart 2. These values again became elevated and she had a recurrence of her symptoms when she was temporarily placed on saline placebos. When the ACTH was reinstituted she again experienced relief of her symptoms and the values returned toward normal. The ACTH was gradually withdrawn over a period of 2 wk. after which it was discontinued for 3 mo. During this time she remained completely free of symptoms and was able to perform physically as a normal child.

Patient M. P.: This 13 yr. old female was 1 of 2 siblings that developed acute rheumatic fever following an ear infection 3 wk. previous to admission to the hospital. She had fever, polyarthritis and carditis at the time of admission. The polyarthritis and fever subsided within 4 and 12 hr.,

respectively, after the institution of ACTH therapy. There was a rapid drop of the sedimentation rate and the hyaluronidase inhibitor and a slow drop of the mucoprotein as seen in chart 3. This patient illustrates the typical response of a patient with acute rheumatic fever to ACTH therapy. Two months after the patient's admission to the hospital, all evidence of carditis had disappeared.

Patient W. P.: This 9 yr. old boy had acute rheumatic fever and diabetes mellitus, the diabetes being present 7½ yr. before the onset of his rheumatic fever. At the time of admission to the

FIG. 1. Patient E. R., 13 yr. old girl with rheumatoid arthritis bedridden for 4 yr. a. Photograph taken just preceding administration of ACTH. Note emaciation, apprehension and limitation of joint movement with muscle shortening. b. Photograph taken 1 mo. later showing weight gain, change in disposition and return of joint movement to normal.

hospital he had fever, carditis and severe polyarthritis requiring a cradle over his extremities to protect his painfully swollen joints. ACTH gave him prompt relief of his arthralgia within 4 hr. and his temperature returned to normal within 24 hr. The response of the mucoproteins, inhibitor and sedimentation rate to the ACTH can be seen in chart 4. Attempts to discontinue the drug on the 18th day of observation were apparently premature as he experienced a recurrence of his symptoms and the sedimentation rate and hyaluronidase inhibitor again became elevated. It can be noted that the mucoproteins were still markedly elevated on the 18th day of observation when the

drug was discontinued. With reinstitution of ACTH therapy the patient again experienced relief of symptoms and the values returned to normal. The elevation of the sedimentation rate on the 50th day of observation was probably due to an influenza type A infection he had at the time along with other children on his ward.

Before ACTH was begun, the insulin requirement of Patient W. P. was approximately 40 units/day. At the time he was receiving ACTH 75 mg./day his insulin requirement was 80 u./day.

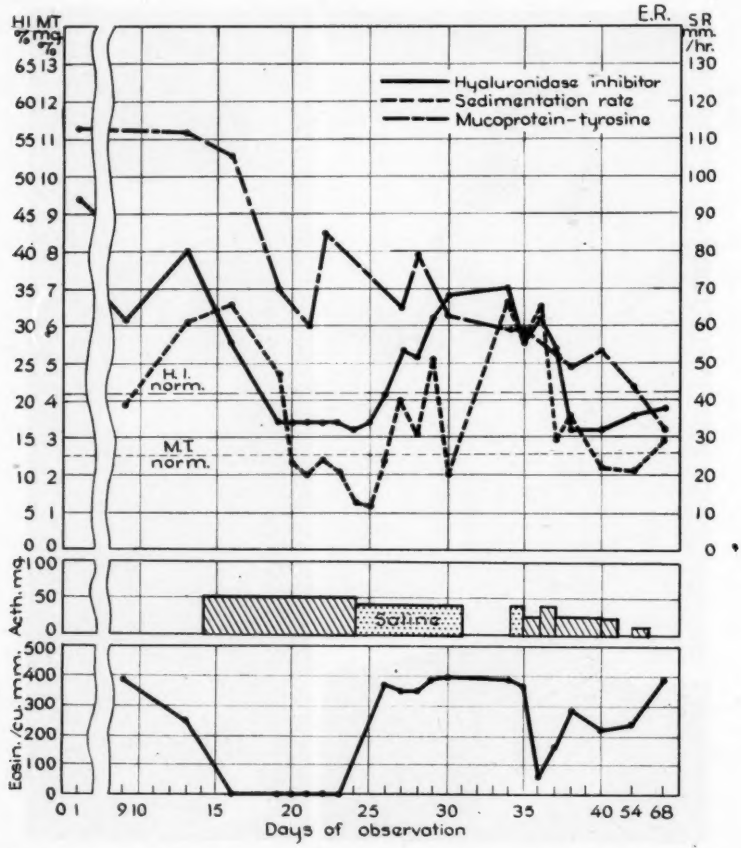

CHART 2. E. R. (see Fig. 1). Note rapid drop of hyaluronidase inhibitor and sedimentation rate but slow drop of mucoproteins during ACTH administration. Off ACTH these values again became elevated and patient's symptoms recurred temporarily but disappeared when ACTH was again started.

With the gradual reduction of the dosage of ACTH, the insulin requirement also decreased. There were no obvious permanent effects of the ACTH on this patient's diabetes mellitus.

Patient E. W.: This was a 17 yr. old female who had had 6 previous episodes of acute rheumatic fever. At the time of her admission to the hospital she had acute rheumatic fever manifested by mild polyarthritis, fever and carditis. Upon receiving ACTH she experienced relief of her symptoms, but her sedimentation rate was slow to return to normal as seen in chart 5. This was the second patient given ACTH by the present investigators and there was therefore some uncertainty about the proper dosage. Because of the persistent elevated sedimentation rate, the dose of ACTH

was gradually increased but the sedimentation rate remained essentially unchanged until additional vitamin C (500 mg./day) was given. Following this the sedimentation rate became normal. However, the supply of ACTH became temporarily exhausted, and in spite of increasing amounts of vitamin C up to 1,000 mg./day, the patient developed a recurrence of her previous symptoms and pericarditis with a friction rub. When ACTH again became available on the 32nd day of observation, she experienced prompt relief with small daily doses and the hyaluronidase inhibitor, mucoprotein

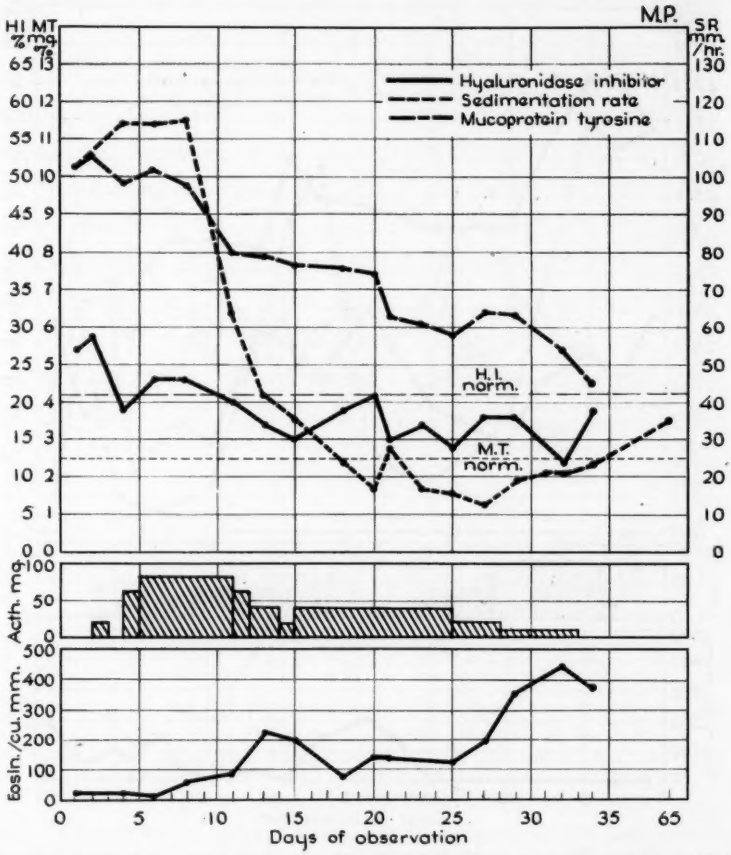

CHART 3. Patient M. P., 13 yr. old female with acute rheumatic fever, polyarthritis and carditis. This shows typical response of hyaluronidase inhibitor, mucoproteins, sedimentation rate and eosinophil count to administration of ACTH.

and sedimentation rate became normal. This patient illustrates the effect of inadequate amounts of ACTH given initially.

Patient P. B.: This was a 12 yr. old female with acute rheumatic fever; polyarthritis, fever and carditis. With the administration of ACTH her symptoms promptly disappeared. Her case is included to show what is interpreted to be the effect of inadequate amounts of ACTH. As can be seen in chart 6, the sedimentation rate and hyaluronidase inhibitor were slow to return to normal. Only when the dose of ACTH was increased markedly on the 32nd day, did the sedimentation rate drop promptly to normal.

Patient P. A.: This was a 13 yr. old boy with subacute rheumatoid arthritis. On admission to the hospital he was not acutely ill but he had the typical swollen, spindle-shaped joints, there being slight pain and slight limitation of movement of these joints. His spleen was just palpable below the costal margin, and he had numerous subcutaneous nodules over the extensor surfaces of his extremities. He had no fever or leucocytosis. Administration of ACTH produced a change to

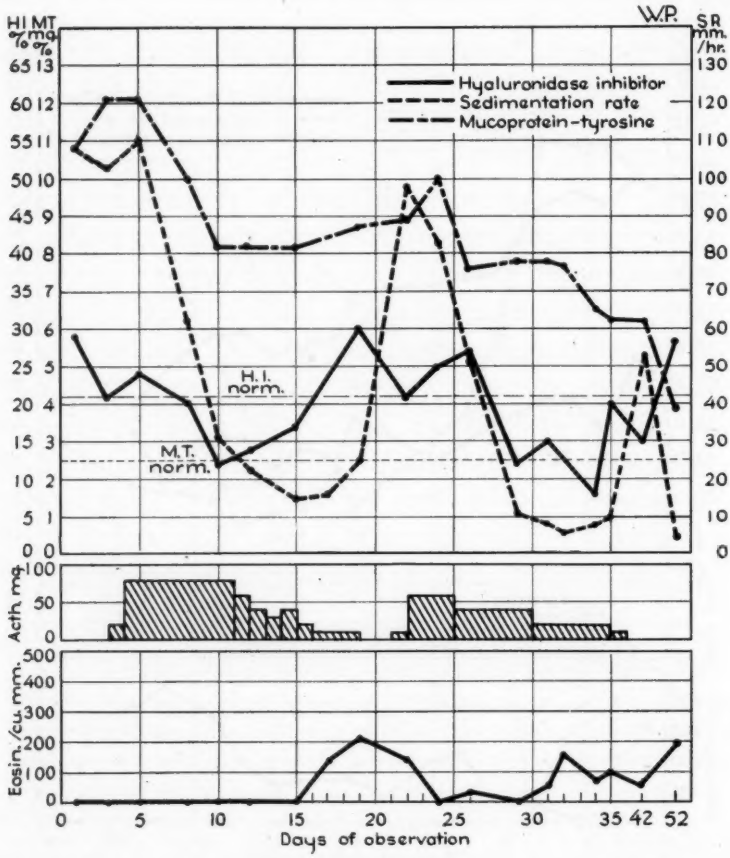

CHART 4. Patient W. P., 9 yr. old boy with acute rheumatic fever, severe polyarthritis, carditis and diabetes mellitus. Note rapid response to ACTH followed by recrudescence when attempts to discontinue drug were premature.

near normal in the appearance of the joints, a reduction to normal in the size of the spleen and a marked reduction in the number and size of the subcutaneous nodules. Initially ACTH produced a drop in the sedimentation rate. Upon further administration of the drug, the values began to rise. As mentioned previously this response has been observed in hypoglycemic children receiving ACTH.[19] After discontinuing ACTH the patient remained relatively free from his presenting complaints and physical pathology.

After the experiments described in this communication were completed, reports appeared by Hakanson and Luft[24] and Schmith and Faber[25] on the effect of ACTH on the

serum hyaluronidase inhibitor and sedimentation rate in patients with rheumatoid arthritis. Each of these reports dealt with only one patient, but their observations are in accord with those herein presented. Subsequently, Dorfman and Moses[26] reported similar results on four children with chronic rheumatic fever.

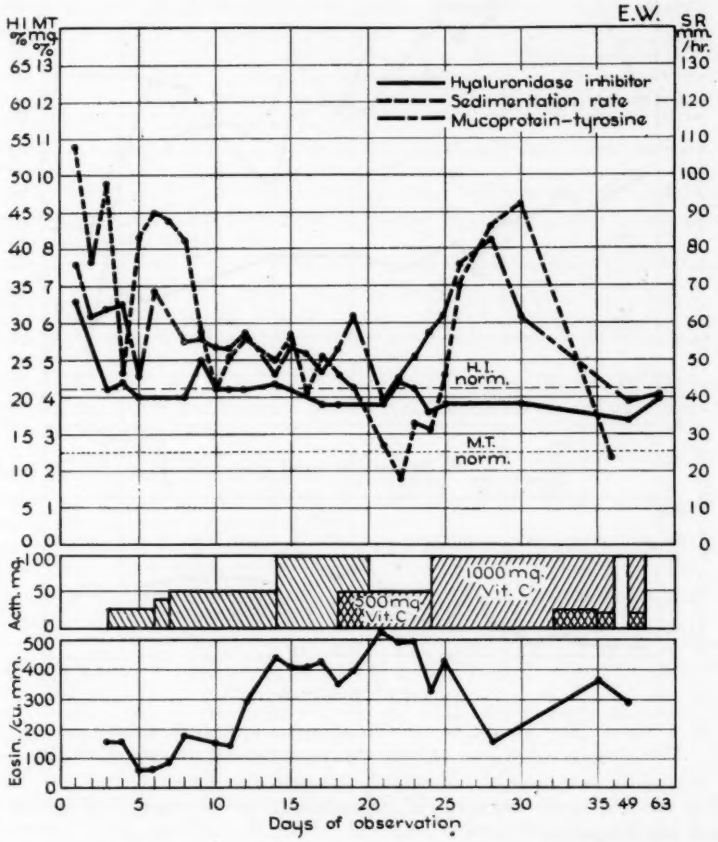

CHART 5. Patient E. W., 17 yr. old female who was experiencing her 6th recurrence of acute rheumatic fever with polyarthritis and carditis. Note slow drop in sedimentation rate probably due to insufficient amounts of ACTH. Added vitamin C was accompanied initially by further drop in SR but without ACTH values again became elevated.

SUMMARY

ACTH was administered to 13 patients with acute rheumatic fever and to two with rheumatoid arthritis. The following observations were made:

1. The elevated nonspecific hyaluronidase inhibitor concentration of the serum dropped rapidly, usually to normal values, within 10 to 15 days, and subsequently the values became subnormal. This seemed to follow closely the subjective improvement in the patient's condition.

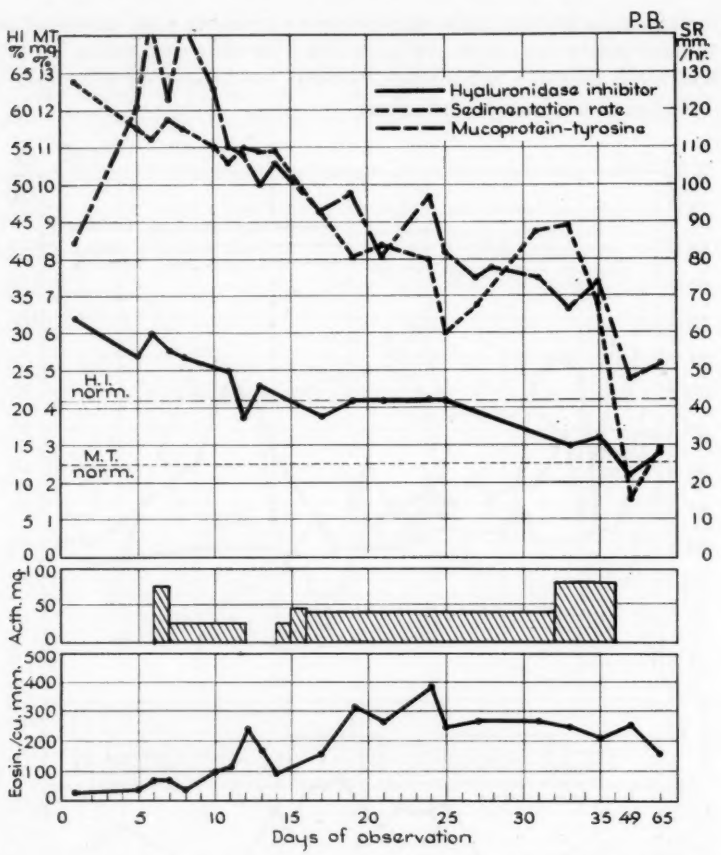

CHART 6. Patient P. B., 8 yr. old female with acute rheumatic fever, polyarthritis and carditis. Slow drop in values was probably due to inadequate amounts of ACTH.

2. The elevated serum mucoprotein concentrations slowly returned to normal over a period of 3 to 4 months.

3. In instances in which the ACTH was withdrawn, or in which the dosage was reduced before a substantial drop of the mucoprotein level had taken place, the disease recurred.

4. ACTH administered to a patient with combined diabetes mellitus and rheumatic fever effected an increase in the insulin requirement to approximately double that of the pre-ACTH period. As the ACTH was gradually withdrawn, the insulin need returned to the pre-ACTH amount. The effect of ACTH on this patient's rheumatic fever was similar to the others receiving the hormone.

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SPANISH ABSTRACT

Efectos de la Hormona Adrenocorticotropica (ACTH) Sobre las Concentracioness de "Hyaluronidase Inhibitor" y "Mucoproteinas" del Suero en las Enfermedades Reumáticas; Sistemas de Enzimas Mucoliticas XV

ACTH fué administrado a trece pacientes con fiebre reumática aguda y a dos con artritis reumátoidea. Las siguientes observaciónes fueron hechas:

1. Una caída rápida de las concentraciones elevadas de "nonspecific hyaluronidases inhibitor" del suero, usualmente a los valores normales en los primeros 10 a 15 días de tratamiento y a valores subnormales subsequentemente. Esta caída de la "hyaluronidase inhibitor" del suero correspondió intimamente con la mejoría subjectiva del paciente.

2. Las concentraciones elevadas de mucoproteinas del suero volvieron lentamente a los valores normales en un periodo de más de tres o cuatro meses.

3. La suspensión o reducción de la dosis de ACTH antes de la ocurrencia de una caída substancial de las concentraciones de mucoproteinas del suero resultó en la recurrencia de la enfermedad.

4. La administración de ACTH a un paciente con diabetes mellitus y fiebre reumática, causó un aumento en las necesidades de insulina aproximadamente el doble de la dosis empleada antes del tratamiento con ACTH. Conjuntamente con la disminución gradual de la dosis de ACTH, las necesidades de insulina del paciente volvieron a las inicialmente empleadas antes del tratamiento con ACTH. Los efectos de la droga en la fiebre reumática de este paciente fueron similares a los obtenidos en los otros pacientes tratados.

University of Minnesota Medical School

STUDIES ON THE LABORATORY DIAGNOSIS OF CYSTIC FIBROSIS OF THE PANCREAS

Positive Gelatin Film* Tests Due to Gelatin-liquefying Bacteria in Feces and Duodenal Juice

By DOUGLAS E. JOHNSTONE, M.D., AND ERWIN NETER, M.D.
Baltimore, Md., and Buffalo, N.Y.

THE laboratory diagnosis of cystic fibrosis of the pancreas is commonly based on the demonstration of the absence or marked decrease of trypsin in the duodenal juice[1] and/or the feces. Shwachman[2, 3] described a simple and rapid method for determination of the presence of trypsin based on the liquefaction of gelatin film. Although he considered the possibility that gelatin liquefaction of gelatin film by fecal specimens may be due to the action of gelatin-liquefying bacteria, he expressed the opinion that the presence of such micro-organisms would not seriously interfere with the reliability of this test at fecal dilutions of 1:5 and 1:10. In a study on the action of various gelatin-liquefying bacteria, Johnstone[4] demonstrated that positive gelatin film tests are readily obtained with broth cultures of gelatin-liquefying bacteria (certain members of the genera Proteus and Pseudomonas) and their Seitz filtrates. It was also shown that the addition of such cultures to trysin-free fecal specimens will give a positive gelatin film test. It is conceivable, then, that the presence of such micro-organisms in the intestinal tract of infants may be responsible for "false"† positive gelatin film tests. To differentiate gelatin liquefaction by pancreatic trypsin from that by gelatin-liquefying bacteria, experiments with enzyme inhibitors were carried out by Johnstone and Neter.[5] It was shown that soy bean trypsin inhibitor does not inhibit gelatin liquefaction by cultures of Proteus and Pseudomonas and their Seitz filtrates.

With these considerations in mind it was deemed of interest to determine on patients with cystic fibrosis of the pancreas (1) the incidence of gelatin-liquefying bacteria in the feces, duodenal juice and throat, (2) the incidence of "false" positive gelatin film tests, (3) the possible value of enzyme inhibitors in this test and (4) the effects of penicillin therapy on the incidence of "false" positive tests. Since an adequate number of cases of cystic fibrosis of the pancreas were not available for study at the Buffalo Children's Hospital, Dr. Dorothy H. Andersen of The Babies Hospital, New York, made it possible for the authors to study many of her patients with this disease as well as her records of many others.

MATERIAL AND METHODS

The gelatin film test on fecal specimens was carried out as described by Shwachman[3] except

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* Gelatin film refers to the roentgenogram film used in routine practice.

† For the sake of brevity, the term "false" positive gelatin film test is used here to connote a positive test probably due to bacterial gelatin-liquefying enzyme rather than pancreatic trypsin.

that the liquefied gelatin film was washed off at the end of the test without gentle rubbing and the material was tested in higher dilutions. The tests with the trypsin inhibitor were carried out as described recently.[5] In brief, the fecal suspension was mixed in Wassermann tubes with equal amounts of soy bean trypsin inhibitor[6, 7] (Worthington Biochemical Company, Freehold, N.J.) containing 2 mg./cc., and physiologic saline as a control. The inhibitor was kept in a sterile stoppered flask under refrigeration when not in use. A drop of each mixture is used for the gelatin film test, which is placed in a covered Petri dish to prevent drying. Duodenal juice was studied in like manner.

For these investigations 78* patients suffering from cystic fibrosis of the pancreas, ranging in age and size from a 2 wk. old, 1.5 kg. premature baby to an 18 yr. old adult, were available for study. In addition, the records of 254 patients,† previously diagnosed as having cystic fibrosis of the pancreas, were studied.

Results

In order to determine how frequently fecal specimens from proved cases of cystic fibrosis of the pancreas yield positive gelatin film tests, the feces of 78 patients were studied. The results are summarized in table 1. It may be seen that positive gelatin film tests were obtained in a fecal dilution of 1:5 in 62% of these patients and in a dilution of 1:10 in 42%. In addition, review of the records of 254 patients† suffering from this disease revealed that of 131 fecal gelatin film tests done on 59 patients 71 (54%) were positive. Of these 59 patients, 34 (57.5%) at some time had positive tests. All had not received pancreatin for at least three days prior to testing.

TABLE 1

Incidence of Positive Gelatin Film Tests on Feces in Various Dilutions in 78 Patients* with Cystic Fibrosis of Pancreas

Dilution of fecal suspension	No. of patients
1:5	48 (62%)
1:10	33 (42%)
1:20	16 (21%)
1:25	9 (12%)
1:50	8 (10%)
1:100	2 (3%)
Total no. of patients with positive gelatin film tests	48 (62%)
Total no. of patients with negative gelatin film tests	30 (38%)
Total no. of patients tested	78 (100%)

* Pancreatin therapy had been withheld for a minimum of 3 days prior to the collection of fecal specimens for testing in every patient.

On the assumption that these "false" positive gelatin film tests are due to the action of gelatin-liquefying bacteria, the incidence of these micro-organisms in the feces of these patients was determined. The data are summarized in table 2. It may be seen that

* Seventeen of these were from the Buffalo Children's Hospital, 54 from the Babies Hospital, New York, 5 from the New York Hospital, 1 from Bellevue Hospital and 1 from the Jewish Hospital of Brooklyn.

† Thirty from Buffalo Children's Hospital, 176 from Babies Hospital, 34 from the New York Hospital, 3 from Bellevue Hospital, and 11 from The Jewish Hospital of Brooklyn. All had been off pancreatin therapy a minimum of 3 days and had been diagnosed clinically and by duodenal intubation.

98% of the patients, whose stool specimens gave positive tests and were cultured, harbored gelatin-liquefying bacteria in their feces. On the other hand, 93% of the patients whose stool specimens did not liquefy gelatin harbored only non-gelatin-liquefying organisms in their feces. The species of gelatin-liquefying bacteria encountered in the

TABLE 2

INCIDENCE OF GELATIN-LIQUEFYING BACTERIA IN FECES OF 78 PATIENTS WITH CYSTIC FIBROSIS OF PANCREAS

	No. of patients	No. of patients cultured	No. of patients with gelatin-liquefying bacteria in feces	No. of patients with only non-gelatin-liquefying bacteria in feces
Patients with positive gelatin film tests	48 (62%)	45	44 (98%*)	1 (2%*)
Patients with negative gelatin film tests	30 (38%)	30	2 (7%)	28 (93%)

* Per cent of patients cultured.

TABLE 3

INCIDENCE OF POSITIVE AND NEGATIVE GELATIN FILM TESTS ON 94* PATIENTS WITH CYSTIC FIBROSIS OF PANCREAS PRIOR TO AND FOLLOWING PENICILLIN THERAPY

	Stool gelatin film tests BEFORE penicillin therapy		Stool gelatin film tests DURING or FOLLOWING penicillin therapy	
	Positive	Negative	Positive	Negative
No. of tests	6 (4 patients)	58 (38 patients)	93 (55 patients)	25 (25 patients)
Total	64 tests on 42 patients		118 tests on 80 patients	

* Some of the data for this table was obtained from the hospital records of patients not personally studied by the authors. However, all had had gelatin film tests for trypsin performed on their feces both before and within 2 or 3 days following institution of penicillin therapy.

feces were as follows: P. ammoniae and other members of this genus were recovered from the feces of 35 patients, Ps. aeruginosa (B. pyocyaneus) from 29 patients and Esch. cloacae from one. Some specimens contained more than one species of gelatin-liquefying bacteria.

It has been shown[8] that the administration of penicillin, particularly when administered by the aerosol route, may be followed by the appearance of, or increase in the number of, gram-negative bacilli in the upper respiratory tract. Some of these gram-negative bacteria may be gelatin-liquefying and can pass the stomach barrier and appear in the feces. It seemed of interest, therefore, to determine whether treatment of patients with cystic fibrosis of the pancreas with this antibiotic may be associated with an increase in the incidence of "false" positive gelatin film test. The findings obtained in the analysis of the available data from both the patients in this series as well as the records reviewed substantiate this assumption, as may be seen from table 3.

That gelatin-liquefying bacteria may be present also in duodenal juice of patients with cystic fibrosis of the pancreas and be responsible for "false" positive gelatin film tests is evident from the fact that such micro-organisms were cultured from 16 of 33 specimens studied bacteriologically. Ps. aeruginosa was found in 14 specimens, P. ammoniae in two,

TABLE 4

INCIDENCE OF POSITIVE GELATIN FILM TESTS AND GELATIN-LIQUEFYING BACTERIA IN DUODENAL JUICE OF 15 PATIENTS WITH CYSTIC FIBROSIS OF PANCREAS*

Highest dilution giving positive gelatin film test	No. of specimens	No. of specimens with gelatin-liquefying bacteria	No. of specimens with only non-gelatin-liquefying bacteria	No. of specimens not cultured
1:1	1	0	1	0
1:5	2	2	0	0
1:10	5	5	0	0
1:20	1	1	0	0
1:25	1	1	0	0
1:50	2	1	0	1
1:100	1	0	0	1
Negative gelatin film test in a 1:1 dilution	2	0	2	0
Total	15	10	3	2

* None of these 15 patients was on pancreatin therapy.

TABLE 5

RELATIONSHIP BETWEEN PRESENCE OF GELATIN-LIQUEFYING BACTERIA IN THROAT AND FECES OF 66 PATIENTS WITH CYSTIC FIBROSIS OF PANCREAS AND POSITIVE GELATIN FILM TESTS ON FECES AND PENICILLIN THERAPY

	No. of patients
(1) Gelatin-liquefying bacteria in throat	66
(1a) Patients in Group (1) subsequent to penicillin therapy	29
(1b) Patients in Group (1) prior to penicillin therapy or without it	11
(1c) Patients in Group (1) subsequent to penicillin therapy who had no pre-therapy throat culture	26
	(66)
(2) Patients in Group (1) with gelatin-liquefying bacteria in feces	28 (29 tested)
(3) Patients in Group (1) with positive gelatin film test on feces	27 (28 tested)

a gelatin-liquefying Chromobacterium in one and Esch. cloacae in one. As may be seen from table 4, duodenal juice contaminated with gelatin-liquefying bacteria gave positive gelatin film tests in dilutions of 1:10 in 10 of the 15 specimens studied.

Table 5 summarizes the data on (1) the presence of gelatin-liquefying bacteria in the

throat, (2) their incidence in the feces, (3) the incidence of positive fecal gelatin film tests and (4) the relationship between the presence of gelatin-liquefying bacteria in the throat and feces to penicillin therapy. It may be noted that almost all fecal specimens tested from patients harboring gelatin-liquefying bacteria in the throat and feces gave positive gelatin film tests and that such positive tests were encountered more frequently in penicillin-treated than in untreated patients. These data further substantiate the impression that these gelatin-liquefying bacteria may be responsible for the apparently "false" positive gelatin film tests.

Discussion

Although the gelatin film test on fecal specimens as described by Shwachman[2, 3] may be a useful tool for the laboratory diagnosis of cystic fibrosis of the pancreas, it is evident from the previous observations[4, 5] and the data presented in this communication that gelatin-liquefaction by bacteria present in the feces or duodenal juice may be responsible for "false" positive results. This impression is based on the above reported close relationship between the incidence of gelatin-liquefying bacteria in feces and duodenal juice and positive gelatin film tests in patients with cystic fibrosis of the pancreas. Penicillin therapy, as reported here, appeared to increase the occurrence of gelatin-liquefying bacteria and, therefore, to increase the incidence of "false" positive gelatin film tests. Attempts to differentiate between the two enzyme systems under consideration, namely, pancreatic trypsin and bacterial gelatinase, were partially successful. It could be shown that soy bean trypsin inhibitor inhibits trypsin but not the bacterial gelatinase. Studies are under way to develop a satisfactory gelatinase inhibitor which will not inhibit trypsin. It is worthy of note that the enzyme responsible for apparently "false" positive gelatin film tests is usually not present in high concentrations. Only 2 of the fecal specimens of 48 patients gave "false" positive gelatin film tests in a dilution of 1:100. In contrast, fecal specimens of normal children of all ages[9] usually gave positive fecal gelatin film tests in a dilution of 1:100. It is apparent, therefore, that a titration of the fecal specimen is of some value.

Of the other possible causes of a positive gelatin film test in the absence of trypsin, the most frequently encountered was the ingestion of pancreatin or similar substances. The presence of gelatin-liquefying bacteria or their enzymes in the diluent used for the preparation of the fecal suspension may also be a cause. It is possible that additional factors may be responsible for false positive gelatin film tests. On the other hand, deficiency of pancreatic trypsin in fecal specimens does not necessarily indicate the diagnosis of cystic fibrosis of the pancreas, since lowering of the trypsin level in feces may be due to functional or other changes of the pancreas or intestinal tract (marasmus,[10] obstipation, megacolon, obstruction of the pancreatic duct by a stone, valve or tumor, etc.). From the preceding data a tentative outline for the study of gelatin-liquefying enzyme in feces and/or duodenal juice of patients suspected of suffering from cystic fibrosis of the pancreas may be proposed.

Recommended Modification of Shwachman Method

The gelatin film test is carried out as described by Shwachman on a fecal suspension in physiologic saline in a dilution of 1:5 and 1:10. A negative test makes further examinations of that specimen unnecessary, although it is advisable to repeat the examination on subsequent specimens. If the gelatin film liquefaction test is positive, the following

tests may be carried out: 1. The fecal specimen should be cultured for the presence of gelatin-liquefying bacteria. If a detailed bacteriologic examination is not feasible, a small amount of the fecal specimen may be seeded into a tube of nutrient gelatin. Together with a control, this tube is then incubated and tested for gelatin liquefaction. 2. Serial dilutions of the fecal suspension are then examined by means of the gelatin film test. A titer of 1:100 or above in the vast majority of cases indicates pancreatic tryptic activity. 3. A suspension of twice the minimal concentration of the specimen yielding a strongly positive gelatin film test is then mixed with equal amounts of (a) soy bean trypsin inhibitor solution containing 2 mg./cc. and (b) physiologic saline as a control. Complete inhibition of gelatin liquefaction by the specimen containing trypsin inhibitor in the presence of a positive control indicates the presence of trypsin. Similar tests may be carried out with duodenal juice. Pancreatin therapy should be withheld for a period of at least three days prior to carrying out these tests, and it is evident from the data just presented that false positive tests may be anticipated far more frequently in patients who were given massive penicillin therapy, due to the possible conversion of the usual bacterial flora of the throat to a flora of gram-negative, gelatin-liquefying bacteria.

Summary and Conclusions

A study of 78 patients with cystic fibrosis of the pancreas was carried out to determine whether gelatin-liquefying bacteria may be responsible for gelatin liquefaction of gelatin film by feces and duodenal juice. In addition, the records of 254 children with cystic fibrosis of the pancreas were reviewed relative to the incidence of apparently false positive gelatin film tests. The following data were obtained:

1. Fecal specimens of 48 out of 78 patients yielded positive gelatin film tests. Only 9 of these 48 specimens were positive in a dilution of 1:25 or higher, but 33 gave positive tests in a dilution of 1:10. Likewise, a review of 254 charts of proved cases of cystic fibrosis revealed that of 131 tests on 59 patients, 71 or 54% were positive.

2. Gelatin-liquefying bacteria were present in the feces of 44 out of 45 patients whose stool specimens gave a positive gelatin film test and which also were examined bacteriologically, but in only 2 out of 28 whose feces gave negative gelatin film tests.

3. Treatment with penicillin may be associated with an increase in the percentage of positive gelatin film tests. Of 42 patients whose feces were obtained prior to or without penicillin therapy only 4 gave positive gelatin film tests, whereas the feces of 55 of 80 patients who had received treatment with penicillin did so.

4. Soy bean trypsin inhibitor partially or completely prevented gelatin liquefaction by fecal specimens of patients with cystic fibrosis of the pancreas only if the latter were diluted to 1:5 or less, possibly indicating the presence of small amounts of trypsin. This titer of soy bean trypsin inhibitor-neutralizable enzyme is far below normal.

5. Gelatin-liquefying bacteria were found in all 10 of the cultured duodenal juice specimens which gave apparently false positive gelatin film tests in a dilution of 1:10 or higher.

6. The incidence of gelatin-liquefying bacteria in the throat and feces of patients giving apparently false positive fecal gelatin film test was far greater in the penicillin-treated patients than in untreated patients.

7. The importance of the apparently false positive gelatin film test is discussed and modifications of this test are outlined which make for greater reliability.

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SPANISH ABSTRACT

Estudio del Diagnóstico de Laboratorio de la Enfermedad Cistica del Pancreas; Pruebas Positivas del "Gelatin Film"* Debidas a las Propiedades Liqueficantes de las Bacterias Existentes en las Heces Fecales y Jugo Duodenal

Se hizo un estudio en 78 pacientes padeciendo de la enfermedad cistica del pancreas, con el proposito de determinar si las propiedades liqueficantes de las bacterias contenidas en las heces fecales y jugo duodenal son las responsables en producir la liquefaccion de la gelatina de las placas radiograficas corrientes, (gelatin film test). Además, se revisaron las historias clinicas de 254 niños con enfermedad cistica del pancreas, en relación a la incidencia de reacciones positivas falsas del "gelatin film test." La siguiente información fué obtenida:

1. De un total de 78 pacientes, 48 tuvieron muestras fecales positivas con la placa radiografica (gelatin film test). Solamente nueve de las 48 muestras fecales fueron positivas en diluciones de 1:25 o mayores, pero 33 muestras fueron positivas en diluciones de 1:10. Similarmente, una revisión de 254 historias clinicas de casos probados de enfermedad cistica del pancreas revelaron que de 131 pruebas realizades en 59 pacientes, en 71 pacientes (54%) fueron positivas.

2. Se observaron bacterias con propiedades liqueficantes de la gelatina, en 44 muestras fecales obtenidad de 45 pacientes, las cuales fueron positivas con el "gelatin film test" y además examinadas bacteriologicamente. De 28 pacientes con muestras fecales negativas con el "gelatin film test," solo en dos muestras se encontraron bacterias con propiedades liqueficantes de la gelatina.

3. Se observó que el tratamiento de estos pacientes con penicilina puede producir un aumento en los porcentajes de pruebas positivas de los "gelatin film test." De 42 pacientes cuyas muestras fecales fueron obtenidas antes del tratamiento con la penicilina, solo 4 muestras fueron positivas con el

* Se refiere a las placas radiograficas en uso corriente.

"gelatin film test," mientras que de 80 pacientes recibiendo penicilina, 55 muestras fueron positivas.

4. Diluciones de muestras fecales obtenidas de pacientes con enfermedad cistica del pancreas, con substancias inhibidoras de la tripsina (soy bean trypsin inhibitor), unicamente evitó la liquefaccion parcial o total de la gelatina, cuando la dilución de la muestra fecal fué de 1:5 o menor, lo que sugiere la presencia de una pequeña cantidad de tripsina. Estos titulos de "soy bean trypsin inhibitor-neutralizable enzyme" estaban marcadamente por debajo de los limites normales.

5. Se encontraron bacterias liqueficantes de la gelatina, en todos los cultivos de muestras de jugo duodenal de pacientes que aparentemente arrojaron pruebas positivas falsas con el "gelatin film" en diluciónes de 1:10 o mayores.

6. La incidencia de bacterias con propiedades liqueficantes de la gelatina, obtenidas de la garganta y heces fecales de pacientes que arrojaron pruebas positivas falsas con el "gelatin film," fué mucho mayor en el grupo tratada con penicilina que en el grupo sin tratar.

7. Se discute la importancia de las pruebas positivas falsas con el "gelatin film" y se presentan modificaciones de esta prueba para una mayor exactitud en los resultados.

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STUDIES IN STEATORRHEA

Effect of the Level of Dietary Fat upon the Absorption of Fat and Other Foodstuffs in Idiopathic Celiac Disease and Cystic Fibrosis of the Pancreas

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IN THE dietary management of states of intestinal intolerance, it has been customary to restrict the type of food to which the intestine was intolerant. Thus, in infantile diarrhea, in which assimilation of all foodstuffs is impaired, it has been the general practice to restrict food as a whole, in pure chronic steatorrheas to restrict fat, and in the celiac syndrome where the assimilation of both fat and carbohydrate are affected to restrict both of these foodstuffs. The basis of this therapy has been: (1) the belief that food intolerance was occasioned by some toxic property of the food, (2) the view that rest to an organ or a function would facilitate recovery and, finally, (3) the fear that abnormal loss of some poorly tolerated food would "wash out" other essential nutrients, thereby impairing rather than aiding nutrition.

In the case of infantile diarrhea the present investigators were led to challenge these traditional concepts. Efforts to implicate toxic components of the food had been singularly unsuccessful. It was shown[1] that recovery from intestinal intolerance was not hastened by the policy of therapeutic starvation. Metabolic studies at different levels of feeding[2] showed no evidence of "washing out" of other nutrients, and even in the severest cases increased intake resulted in increased assimilation of every nutrient studied. These findings confirmed the clinical impressions of the value of a generous oral intake as a supplement[3] to needed parenteral therapy.

These observations in acute diarrhea suggested that the conventional food restrictions in chronic steatorrheas might not necessarily be well founded. Therefore, it was undertaken to study the effect of varying the fat intake in various steatorrheas in order to ascertain the effect of fat administration on fat tolerance and on the assimilation of other foodstuffs. If fat administration were without noxious effects, it was conceivable that excessive losses of fat could be compensated for by increasing the fat intake. Studies which the present authors have carried out on the steatorrhea of the premature infant have been reported previously.[4] Briefly, it was found that the premature infant could tolerate intakes of fat well in excess of those ordinarily given, no untoward effects on fat tolerance being observed. The percentage absorption was not significantly affected by the intake and an increased intake thus served to increase the absolute assimilation.

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The present report deals with the effect of varying the fat intake in two other conditions associated with steatorrhea, namely, "idiopathic" celiac disease and cystic fibrosis of the pancreas. Since these two conditions are essentially different—the former being dependent on a defective assimilatory mechanism in the intestinal wall and the latter upon a deficiency of digestive enzymes—they will be discussed separately.

I. "Idiopathic" Celiac Disease

Some pertinent observations were already in the literature. Macrae and Morris[5] had studied the effect of varying the fat intake in four celiac patients. Increasing the intake from a normal level (24 to 38 gm./day) to a high level (75 to 115 gm./day) produced striking increases in fat absorption, the percentage absorption showing no consistent change; untoward clinical effects were not observed. Similar observations were made by Black, Fourman and Trinder[6] and by Bassett et al.[7] in adult sprue, a condition generally regarded as the adult counterpart of celiac disease. Extensive clinical observations were made by Haas,[8] who maintained celiac patients on a generous fat intake for prolonged periods and concluded that definite clinical improvement could be attributed to this regime. Against these observations are various clinical reports in which relapse is believed to have been initiated by the introduction or increase of fat in the diet. In interpreting such reports it is necessary to bear in mind that genuine relapses with loss of tolerance are common in celiac patients without any change in regimen and that a mere increase in steatorrhea following the administration of fat does not necessarily indicate a relapse; it may mean merely that a defective absorption mechanism is being demonstrated by a load.

The present studies were carried out on three celiac patients admitted to the Children's Medical Service at Bellevue Hospital whose protocols were as follows:

Case 1. W. G. (B.H. 44003-47), an 8 mo. old white boy, was admitted to the hospital because of chronic diarrhea of 6 wk.' duration. At the time of admission he was having 5 to 10 loose, bulky, pale, foul-smelling stools a day. He was a pale, irritable boy showing evidence of marked weight loss. The extremities were wasted and the abdomen was somewhat protuberant. An assay of the duodenal juice showed an adequate concentration of trypsin. The glucose tolerance test gave a flat curve. Stool cultures were repeatedly negative for enteric pathogens. The course in the hospital was stormy. After adequate parenteral therapy he was first placed on the skimmed milk-casec formula he had been receiving at home. One week after admission he developed bronchopneumonia and was slowly recovering when the diarrhea, which had improved somewhat, flared up again. A vigorous course of parenteral fluids followed when all appetite was lost and vomiting ensued. When he failed to gain weight, in spite of recovery of appetite 2 wk. later, he was transferred to the metabolism ward for study. He was placed on a basal diet of evaporated milk, sugar and water, providing 110 cal./kg. and 33.5 gm. fat/day. In subsequent periods this was supplemented by the addition of 30 and later 90 gm. butter/day. Although the stools increased in size and were obviously more fatty, there was no clinical exacerbation in the boy's condition and the weight gradually rose. A supplement of homogenized meat* was added to the high fat diet in a fourth period which induced a further gain in weight. In the succeeding 2 mo. during which he remained on this diet he gained 4.5 kg. He was subsequently allowed to choose his own diet. A follow-up report indicates that his progress has been excellent, his weight being 19.1 kg. at the age of 2½ yr. Except for some protuberance of the abdomen, there is no evidence of the celiac picture. Two relapses occurred after discharge from the hospital, both occurring on the prescribed diet and both definitely related to parenteral infection with fever. The metabolic data is presented in table 1 and the weight curve in chart 1.

Examination of the data reveals that the addition of fat even at the extremely high level caused virtually no drop in the percentage absorption and caused a striking absolute increase in the assimilation of fat. The final period, far from showing any loss of tolerance to fat from the continuation

* Supplied by Swift & Company, New York.

TABLE 1

CASE 1*

		Water (cc./day)	Ash (gm./day)	Na (mEq./day)	K (mEq./day)	Ca (mEq./day)	P (mEq./day)	Cl (mEq./day)	Fat (gm./day)	N (gm./day)	Wt. kg.
Control Diet	Intake	1080	6.70	21.9	33.4	52.5	23.0	24.8	**33.50**	4.62	8.0
	Urine	293	1.92	5.7	24.4	0.3	7.9	23.4		2.45	
	Feces	189	4.00	0.8	4.2	46.5	17.0	1.2	14.20	0.74	
	Absorbed	891	2.70	21.1	29.2	6.0	6.0	23.6	**19.30**	3.88	
	% Intake Absorbed	82	40	99	88	11	26	95	58	84	
Control Diet	Intake	1260	7.32	31.8	34.1	54.0	23.1	34.9	**59.60**	4.62	8.0
+30 gm.	Urine	425	3.06	19.5	29.8	0.3	8.2	31.4		1.85	
Butter/Day	Feces	236	4.07	2.3	4.6	51.0	14.1	2.8	28.50	0.67	
	Absorbed	1024	3.25	29.5	29.5	3.0	9.0	32.1	**31.10**	3.95	
	% Intake Absorbed	81	44	93	87	6	39	92	52	86	
Control Diet	Intake	1050	8.59	51.6	34.3	57.0	23.3	54.8	**111 70**	4.62	8.1
+90 gm.	Urine	446	2.96	26.2	30.0	0.6	6.8	41.5		1.30	
Butter/Day	Feces	293	4.65	5.2	5.8	74.9	21.8	2.1	60.50	0.72	
	Absorbed	757	3.94	46.4	28.5	−17.9	1.5	52.7	**51.20**	3.90	
	% Intake Absorbed	72	46	90	86	−31	6	97	46	85	
Control Diet	Intake	1280	10.62	66.8	49.5	57.3	25.5	60.7	**111.70**	8.88	8.8
+90 gm.	Urine	430	3.80	28.2	36.8	0.2	14.3	44.8		2.42	
Butter+170	Feces	224	5.90	15.7	12.8	51.0	10.8	2.6	34.00	1.46	
gm. meat/day	Absorbed	1056	4.72	51.1	36.7	6.3	14.7	58.1	**77.70**	7.42	
	% Intake Absorbed	83	45	77	74	11	58	96	67	84	

* The metabolism periods in this and other patients here reported were 4 days in duration, stools being marked with carmine The patients were with 1 exception (Case 2) placed on metabolism frames to permit separate collections of urine and feces The analytic methods employed were as described in a previous publication.[3] The basal diets were prepared from evaporated milk and sugar, providing approximately 100 cal./kg. of which approximately 15% of the calories were from protein, 50% from carbohydrate and 35% from fat. The supplementary fat was supplied as melted butter, fed by spoon.

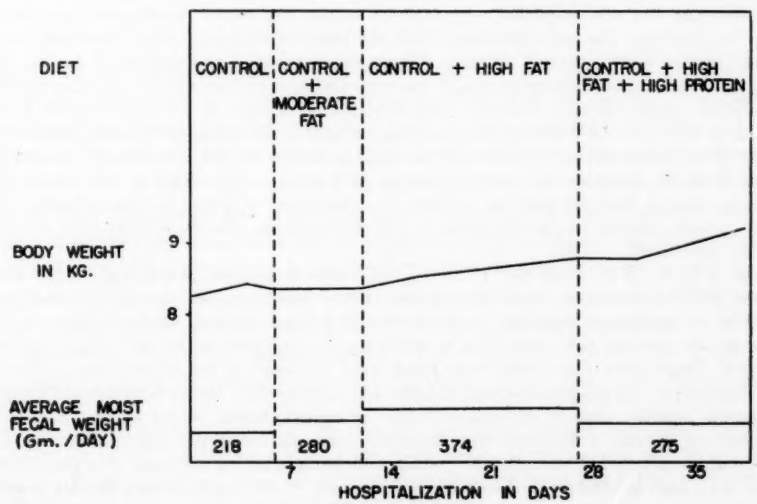

CHART 1. (Case 1)

of this diet, showed the reverse. Little change was noted in the absorption of nitrogen or minerals from the increase in dietary fat, the only exception being a temporary loss of calcium immediately following the institution of the highest fat intake; this was not, however, sustained.

Case 2. R. M. (B.H. 41757-49), a 7½ yr. old white boy, was admitted to the Bellevue Metabolism Ward for study because of large bulky stools and failure to gain properly in weight and height since 6 mo. of age. The history from another hospital indicated that at the age of 4 mo. he began to have intermittent diarrhea, the stools being reported as large, foul and bulky at 10 mo. when he was hospitalized. The birth weight was not quite doubled at that time although the mother claimed the child's appetite was excellent. Stool cultures were negative for enteric pathogens. The glucose tolerance curve was flat. The course after discharge from the hospital was uneventful except that the weight gain was slow as was the gain in height. The parents were of normal height and weight.

TABLE 2

CASE 2

		Water (cc./day)	Ash (gm./day)	Na (mEq./day)	K (mEq./day)	Ca (mEq./day)	P (mEq./day)	Cl (mEq./day)	Fat (gm./day)	N (gm./day)	Wt. kg.
	Intake	1920	14.84	50.0	79.0	168.0	52.6	46.0	**76.80**	8.06	20.6
	Urine	525	5.40	33.3	48.4	0.6	12.5	25.8		2.98	
Control Diet	Feces	186	5.35	4.7	7.0	81.5	19.1	0.9	41.50	1.53	
	Absorbed	1734	9.49	45.3	72.0	86.5	33.5	45.1	**35.25**	6.53	
	% Intake Absorbed	90	64	91	91	52	64	98	46	81	
Control Diet	Intake	1920	16.64	78.2	79.8	172.3	52.9	74.8	**153.60**	8.06	20.6
+85.4 gm.	Urine	679	7.26	48.6	45.5	0.7	12.8	40.5		3.38	
Butter/Day	Feces	272	6.50	8.2	9.2	84.5	21.0	2.1	89.40	1.54	
	Absorbed	1648	10.14	70.0	70.6	87.8	31.9	72.7	**64.20**	6.52	
	% Intake Absorbed	86	61	90	89	51	60	97	42	81	

The abnormal stools persisted but became less frequent with time. When seen in this clinic at 7½ yr. of age, he was 111 cm. in height and weighed 20.0 kg. He was a well proportioned, small, somewhat thin boy who resembled a 6 yr. old in stature. The mental development was consistent with the actual age. The stools were large, foul and fatty, numbering not more than 3/day. Following the method of Shwachman,[9] dilutions of stool samples showed normal tryptic digestion. It was felt that the clinical and laboratory picture was consistent with the diagnosis of the celiac syndrome. The fullest cooperation was secured not only in taking his diet but in the careful collection of his excreta as well. Data on 2 levels of fat are shown in table 2. On discharge from the hospital it was suggested that supplements of fat and protein be given so that the amounts of these foodstuffs consumed would be approximately doubled. It is too early to record the results on this regime.

As is apparent from the table the addition of a supplement of butter fat which doubled the fat intake virtually doubled the fat absorption as well, the percentage absorption being 42% as opposed to 46% of the intake.

Case 3. D. C. (B.H. 46280-49), an 18 mo. old Puerto Rican infant, was brought to the hospital because of fever and a sore mouth. He had been in poor health since the age of 1 yr. when he had the onset of intermittent diarrhea, progressive weight loss and gradual increase in the size of his abdomen. He appeared pale, chronically ill with extreme emaciation and he had a large protuberant abdomen. Large, pale, fatty stools were noted from the onset of his hospitalization. Five stool examinations for ova and parasites were negative and the duodenal contents were repeatedly negative for giardia lamblia. Assay of the duodenal juice for trypsin showed normal activity. A vitamin A tolerance test revealed a flat curve. The causes for admission (fever and sore mouth) were found to be due to infectious mononucleosis, the diagnosis being made on the basis of a positive blood smear and a high heterophile antibody titre. After recovery, he developed a herpes simplex stomatitis. After this had subsided, he was transferred to the Metabolism Ward for study. He was placed on a

purified diet consisting of protein 45 gm., carbohydrate 137 gm. and fat 45 gm. for the control period. The data for this period as well as a period of high fat intake are given in table 3. He had an excellent gain in weight during his stay on the Metabolism Ward amounting to 1.5 kg. in less than 2 mo.

The data presented in the table indicate that this patient, like the preceding 2, was benefited by the fat supplementation. On a high fat diet, he assimilated more fat with apparent clinical benefit. These observations thus confirm those of Macrae and Morris, of Black, Fourman and Trinder and of Bassett et al. as to the effect of increasing fat intake on fat absorption. Pertinent data from the 2 former reports are presented in tables 4 and 5. This experience likewise confirms that of Haas, that generous fat intakes are well tolerated. Such relapses as the present authors have observed have been definitely related to parenteral infection rather than to changes in the diet.

TABLE 3*

Patient D.C. Age 20 mo. Celiac

EFFECT OF INCREASING FAT INTAKE ON FAT ABSORPTION

	Period I	Period II	Period III	Period IV
Fat Intake (gm./day)	**36.0**	**36.0**	**45.0**	**92.7**
Fat in Feces (gm./day)	5.3	8.5	6.7	10.0
Fat Absorbed (gm./day)	**30.7**	**27.5**	**38.3**	**82.7**
Per cent Intake Absorbed	85.1%	76.5%	85.0%	89.1%

* Periods 10 days each.

II. CYSTIC FIBROSIS OF THE PANCREAS

Pancreatic steatorrhea differs from that of "idiopathic" celiac disease in that the defect involves the preparation of food for absorption rather than the process of absorption itself. Both protein and fat digestion are interfered with, carbohydrate being as a rule only minimally affected. The restriction of fat intake has been enthusiastically advocated by Andersen[10–13] in these cases and she has supported this view by chemical observations from which the conclusion was drawn that fat not only increases the steatorrhea, but also exercises a deleterious influence on the nitrogen and mineral balance. An examination of the metabolic evidence presented by Andersen finds it open to criticism, as will be discussed below. Some of it is, moreover, quite at variance with observations of Coffey et al.[14] and of Handelsman et al.[15] on dogs deprived of their pancreatic secretion and with observations on one patient by May and Lowe.* For these reasons it seemed desirable to make further studies of patients with this condition at different levels of fat intake.

* A personal communication from C. D. May describes an unpublished experiment carried out by himself and C. U. Lowe in which a child with cystic fibrosis of the pancreas was fed at 2 levels of fat, e.g., 35 gm. and 85 gm./day. The percentage retention was not affected. On the former diet the absolute absorption was 29.8 gm. and on the latter 73.9 gm.

TABLE 4

	Diet	Fat Intake gm./Day	Fat Excretion gm./Day	Fat Absorbed gm./Day	% Intake Absorbed
Case 1					
	N	29.4	3.77	25.61	87.2
	H	99.2	5.98	93.22	94.2
	N	24.2	7.99	16.21	67.0
	H	97.0	18.53	78.47	80.9
	N	24.0	2.94	21.1	87.6
	H	104.3	12.11	92.2	88.4
Case 2					
	N	34.1	4.45	29.6	87.2
	H	99.9	4.56	95.3	95.4
	N	38.4	12.08	26.4	68.7
	H	75.1	11.37	63.7	84.8
Case 3					
	N	28.2	7.46	20.7	73.6
	H	114.7	22.20	92.7	80.8
Case 6					
	N	38.8	9.91	28.9	74.4
	H	115.0	9.62	105.4	91.6

Data from Macrae and Morris showing the effect of normal (N) and high (H) fat intakes on the absorption of fat in celiac syndrome. Cases 4 and 5 were placed on a single level of fat only.

TABLE 5

ABSORPTION OF FAT IN TROPICAL SPRUE ON 2 LEVELS OF INTAKE

(Data of Black, Fourman and Trinder)

Patient	On 69 gm. Fat/Day		On 96 gm. Fat/Day	
	Fat Absorbed gm.	% Absorbed	Fat Absorbed gm.	% Absorbed
1	56.6	82	79.9	83
2	55.0	80	71.0	74
3	57.3	83	84.5	88
4	58.6	85	83.5	87
5	35.2	51	42.2	44

Studies on three patients with this condition have been carried out and are presented herewith.

Case 4. T. B. (B.H. 5226-48), an 8½ mo. old white boy, was transferred to the Bellevue Metabolism Ward from the Beth Israel Hospital where he had previously been studied. He had a history of a nonproductive cough and of loose, bulky, foul stools which were noted shortly after birth. The diagnosis of cystic fibrosis of the pancreas was established by the lack of trypsin in the

TABLE 6

CASE 4

		Water (cc./day)	Ash (gm./day)	Na (mEq./day)	K (mEq./day)	Ca (mEq./day)	P (mEq./day)	Cl (mEq./day)	Fat (gm./day)	N (gm./day)	Wt. kg.
Control Diet	Intake	900	5.84	20.8	30.8	47.6	25.9	25.4	**27.20**	4.26	7.5
	Urine	175	2.83	7.6	21.5	1.0	9.9	9.2		1.74	
	Feces	87	2.63	0.1	2.0	40.1	9.2	0.4	16.25	0.69	
	Absorbed	813	3.21	20.7	28.8	7.5	16.7	25.0	**10.95**	3.57	
	% Intake Absorbed	90	55	99	94	16	65	99	40	84	
Control Diet +60 gm. Butter/day	Intake	1020	7.10	40.6	31.6	50.6	26.1	45.8	**79.40**	4.26	7.5
	Urine	160	3.42	17.3	29.0	1.1	10.2	21.9		2.04	
	Feces	100	2.37	0.2	1.9	45.2	10.4	0.3	33.20	0.73	
	Absorbed	920	4.73	40.4	29.7	5.4	15.7	45.5	**46.20**	3.53	
	% Intake Absorbed	90	67	99	94	11	60	99	58	83	

duodenal juice. The glucose tolerance and vitamin A tolerance curves were flat. He was a fairly well developed, but poorly nourished boy, showing persistent inspiratory rales over both bases posteriorly. Metabolism periods consisted of a control diet and a test diet in which the fat intake was tripled. The diets were tolerated well and no exacerbation of the diarrhea was induced. The metabolic data are given in table 6.

It will be noted that the added fat, which was the only variable in the diet, produced a striking increase in absolute fat absorption, though a negligible change in the percentage absorption. In contrast to the contention of Andersen, no untoward influence on the absorption of nitrogen or minerals is discernible.

Case 5.* J. H. (145602) was a 3¾ yr. old white boy who was readmitted to Vanderbilt Hospital for the fifth time for metabolic studies. He was first observed at the age of 11 mo. with a history of chronic upper respiratory infections and passage of large, fatty, foul stools since 5 mo. of age. Duodenal drainage showed absence of tryptic digestion. The vitamin A tolerance curve was flat. He continued to have upper respiratory infections but gained weight satisfactorily on a high protein, high carbohydrate, low fat diet with panteric granules. At the time of the metabolism study, his nutrition was good.

TABLE 7

CASE 5

		Water (cc./day)	Ash (gm./day)	Na (mEq./day)	K (mEq./day)	Ca (mEq./day)	P (mEq./day)	Cl (mEq./day)	Fat (gm./day)	N (gm./day)	Wt. kg.
Control Diet	Intake	1883	11.15	92.0	58.0	126.0	41.2	34.4	**61.90**	5.50	14.7
	Urine	1153	4.84	56.5	42.8	1.5	7.6	21.0		3.22	
	Feces	78	4.26	5.7	6.3	62.0	13.3	0.3	24.80	1.07	
	Absorbed	1805	6.89	86.3	51.7	64.0	27.9	34.1	**37.10**	4.43	
	% Intake Absorbed	96	62	94	89	51	68	99	60	80	
Control Diet +112.5 gm. Butter/day	Intake	1815	12.98	98.0	60.5	130.0	41.6	37.2	**163.15**	5.50	14.7
	Urine	900	5.94	68.3	48.7	1.4	6.7	45.7		3.15	
	Feces	248	3.28	5.9	4.0	59.2	13.0	1.4	37.10	1.23	
	Absorbed	1567	9.70	92.1	56.5	70.8	28.6	35.8	**126.05**	4.27	
	% Intake Absorbed	86	75	94	93	54	70	96	77	78	

* For the opportunity to study the Cases 5 and 6, the authors are indebted to Dr. Amos Christie of the Vanderbilt Hospital, Nashville.

Table 7 shows the effect of a 2½ fold increase in the fat intake, which increased strikingly the total fat absorption. No untoward manifestations of a clinical or chemical nature were noted.

Case 6.* D. S. (168046), a 22 mo. old white boy, was admitted to the Vanderbilt Hospital for the second time. His first admission was at the age of 18 mo. when he presented a history of

TABLE 8

CASE 6

		Water (cc./day)	Ash (gm./day)	Na (mEq./day)	K (mEq./day)	Ca (mEq./day)	P (mEq./day)	Cl (mEq./day)	Fat (gm./day)	N (gm./day)	Wt. kg.
Control Diet	Intake	1468	8.15	67.0	43.5	92.0	29.0	25.0	**45.15**	4.02	10.0
	Urine	445	2.26	21.0	21.8	0.6	5.4	2.6		1.20	
	Feces	148	4.05	1.7	9.3	56.2	11.0	0.7	20.80	1.65	
	Absorbed	1320	4.10	65.3	34.2	35.8	18.0	24.3	**24.35**	2.37	
	% Intake Absorbed	90	50	98	79	39	62	97	54	59	
Control Diet +75 gm. Butter/day	Intake	1752	9.37	71.0	45.4	94.7	29.2	25.9	**112.65**	4.02	9.5
	Urine	810	3.65	36.9	29.2	1.3	9.7	7.2		1.38	
	Feces	121	3.66	0.9	5.8	55.0	13.3	1.1	63.00	1.95	
	Absorbed	1631	5.71	70.1	39.6	39.7	15.9	24.8	**49.65**	2.07	
	% Intake Absorbed	93	61	99	87	42	54	96	44	52	

TABLE 9

FAT ABSORPTION IN DOGS DEPRIVED OF EXTERNAL PANCREATIC SECRETION

(Data of Coffey, Mann and Bollmann)

	gm./day			
Fat intake	17.0	36.0	36.0	106.0
Fat in stool	13.4	23.4	18.4	49.8
Fat absorbed	3.6	12.6	17.6	56.2

TABLE 10

ABSORPTION OF N AND FAT IN DOGS WITH LIGATED PANCREATIC DUCT

(Data of Handlesman, Golden and Pratt)

	N		Fat	
	intake (gm.)	absorbed (gm.)	intake (gm.)	absorbed (gm.)
Standard diet	10.4	5.7 (56%)	19.4	11.7 (60%)
High fat diet	10.0	6.3 (63%)	49.1	46.0 (94%)

chronic recurrent respiratory infection, poor weight gain and large, foul, bulky stools from birth. Duodenal enzymes were reported absent. The vitamin A tolerance curve was flat. He was discharged from the hospital on a high protein, high carbohydrate, low fat diet with panteric granules. Penicillin aerosol was given daily. He did fairly well on this regime until 2 days before admission when he developed an upper respiratory infection and was asked to come to the hospital for treatment and study. He was a poorly developed, poorly nourished irritable boy who had numerous rales and

* For the opportunity to study the Cases 5 and 6, the authors are indebted to Dr. Amos Christie of the Vanderbilt Hospital, Nashville.

rhonchi throughout the chest. He was studied on 2 levels of fat intake, one, 2½ times the other. The study was uneventful clinically. The data presented in table 8 again indicate increased absorption of fat at the higher intake and no significant loss in percentage absorption.

It is thus apparent that these studies of cystic fibrosis of the pancreas are in agreement with that of May and Lowe and with those of Coffey, Mann and Bollman as well as those of Handelsman et al. in dogs deprived of their pancreatic secretion, significant data from whose experiments is presented in tables 9 and 10. Not only is fat absorption improved

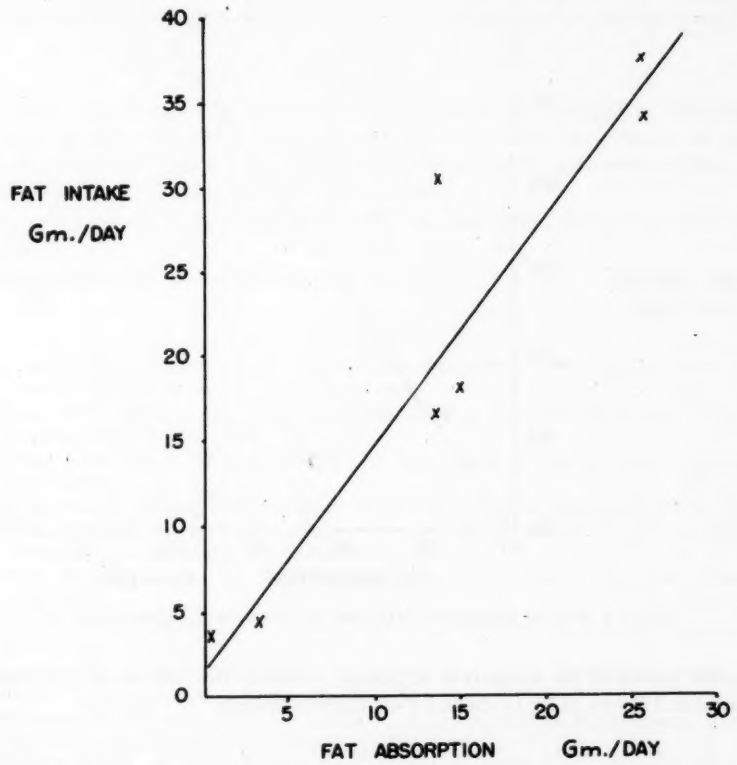

CHART 2. Relation between fat intake and fat absorption. (Data of Andersen.)

by the administration of fat, but no deleterious effect of fat on nitrogen absorption is found.

In examining the data of Andersen[11] it is of interest to note that although fat excretion in the stools was increased in her studies by raising the fat intake, the absorption of fat, not tabulated by her, was also increased. The linear relation between fat intake and absorption is quite as apparent in her studies (chart 2) as in the present report (chart 3).

It has not been possible to find in the data of Andersen evidence for her statement that high fat intakes lead to protein loss by the "cathartic" action of the undigested fat. It is unfortunate that when studying the action of fat, Andersen failed to keep the protein intake constant, and it would seem likely that some of the changes in nitrogen excretion

which she observed were occasioned by changes in nitrogen intake. The variations in mineral excretion which she observed were most irregular and showed no impressive correlation to the fat intake. The present data indicate the constancy of the mineral metabolism in the face of alterations in fat intake, with the one exception of calcium, the absorption of which was adversely, though only temporarily, affected in Case 1. An explanation of the anomalous behavior of calcium in this instance has not been found. Conceivably the patient was deficient in vitamin D, for a D supplement was not given

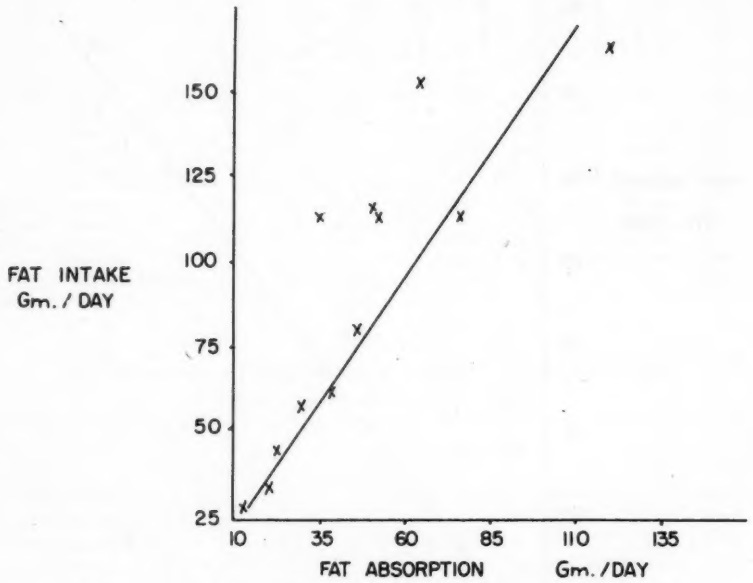

CHART 3. Relation between fat intake and fat absorption. (Authors' data.)

during the period of the study. It is of interest, however, that one of the cases studied by Macrae and Morris (Case I) showed a similar phenomenon.

COMMENT

It is not the purpose of this report to recommend a specific diet for the treatment of the celiac syndrome or of cystic fibrosis of the pancreas. The purpose of the study was to examine the fundamental nature of the metabolic disturbance in these conditions, on which sound dietotherapy must be based.

The authors have examined the evidence supporting the view that dietary fat leads to intolerance to fat and other foodstuffs and have found it wanting. Patients have been subjected to fat intakes far above the normal usage and it was found that these were well tolerated.

The authors do not deny that it is usually possible to meet the nutritional requirements of these subjects by diets poor in fat if sufficient care in the construction of the diet is taken, but one should not lose sight of the fact that fat, even when only 50% of the

intake is absorbed, is still the most efficient source of calories we possess. Interesting knowledge is gradually coming to light on a number of noncaloric functions of fat, a subject recently reviewed by Deuel.[16] Quite apart from the need of some fat to supply essential fatty acids, fats appear to be anti-allergic in nature and there is some evidence that they play a role in immunity. Swift, Forbes and others[17, 18] have shown that they make for more efficient utilization of protein. According to a recent report,[19] a generous fat intake stimulates epiphyseal maturation, particularly in the male, abolishing the lead which the preadolescent female has in this respect. For these reasons it would seem wise to hesitate before avoiding a valuable asset in the management of nutritional problems.

Summary

Metabolic observations were carried out in three patients with "idiopathic" celiac disease and three patients with cystic fibrosis of the pancreas at different levels of fat intake.

It was found that fat absorption remained proportional to fat intake even at high levels. Increasing the intake thus led to improved absorption.

Deleterious clinical or metabolic effects were not encountered with one possible minor exception.

The implications of these observations are discussed.

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SPANISH ABSTRACT

Estudios en Esteatorrea: Los Efectos del Nivel de Grasa de la Dieta sobre la Absorción de Esta y Otros Productos Alimenticios en Pacientes con Enfermedad Celíaca Idiopática y Fibrosis Cística del Pancreas

Observaciones metabolicas fueron realizadas en tres pacientes con enfermedad celíaca idiopática y en otros tres con fibrosis cistica del pancreas.

Se encontró que la absorción de grasa se mantuvo proporcional a la cantidad ingerida en la dieta. Un aumento en la ingestión de grasa fué seguido por una absorción mayor. En general, no se encontraron efectos clínicos o metabolicos nocivos a excepción de una posible alteración insignificante.

Los autores discuten los resultados e implicaciones de estas observaciones.

477 First Avenue

GALACTOSEMIA

Report of Four Cases

By GEORGE N. DONNELL, M.D., AND STANLEY H. LANN, M.D.
Los Angeles

THE clinical syndrome known as galactosemia or galactose diabetes was first described by Von Reuss[1] who, in 1908, presented an 8 week old infant retarded in physical development. Galactose was found in the urine; on a milk-free diet the infant showed marked improvement and the urine became galactose-free.

Since then, a few publications have appeared which establish the syndrome as a definite clinical entity.[2-10] All the patients showed retarded development, hepatomegaly, albuminuria and galactosuria. In many, during the neonatal period, icterus developed which was followed by vomiting, weight loss and the characteristic urinary findings. Most authors agree on the clinical aspects of this disease but there has been a wide difference of opinion concerning the etiology.

The purpose of this paper is to present four additional cases of galactose intolerance. The second case (T. A.) had two siblings with typical clinical findings of early galactosemia; in neither was the disease suspected before death. Case histories of both siblings, with the autopsy findings on one, are included.

CASE REPORTS

Case 1. B. P., a female infant of 15 days, was first admitted to the Los Angeles Children's Hospital for study because of severe vomiting, jaundice and dark urine of 1 wk.' duration. She was the first-born of healthy young parents. Delivery at term was normal and the birth weight was 2640 gm. There was no family history of hereditary or metabolic disorders.

The infant was apparently well for 8 days when she began to vomit persistently; the following day jaundice and dark-colored urine appeared.

Examination revealed a small, vigorous and somewhat dehydrated female infant with evident loss of subcutaneous fat. The skin and sclerae were slightly icteric. Temperature was normal; weight was 2270 gm. Pupils reacted to light and the ocular fundi appeared normal. The lungs and heart were not remarkable. The abdomen was slightly distended and the superficial veins were prominent. The liver was palpable 4 cm. below the costal margin and was firm in consistency. The spleen was not felt.

On admission, Hgb. was 17.9 gm./100 cc., WBC 14.1 thousand/cmm. with a differential of 29% polymorphonuclear leukocytes and 71% lymphocytes. The urine revealed 4+ albumin but was otherwise negative. The icterus index was 70 units and the Coomb's (developing) test negative.

A tentative diagnosis of sepsis was made and the infant was started on penicillin and sulfadiazine. The preliminary blood culture yielded no growth. Urine culture revealed numerous colonies of E. coli and B. aerogenes, so streptomycin was given in addition for 5 days.

For the 1st 5 days in the hospital, the infant was fed a special oral amigen® formula[11] and thereafter an evaporated milk formula of full caloric value. During the entire hospitalization of 4 wk., repeated urine specimens revealed marked albuminuria and a constant but moderate glycosuria without ketonuria. Other significant laboratory data include a NPN of 45 mg./100 cc., a fasting blood sugar of 99 mg./100 cc., a negative Kahn test and negative cephalin flocculation test. Roentgenograms of the long bones, a cystogram and an intravenous pyelogram were within normal limits.

From the Children's Hospital of Los Angeles and the Pediatric Department, School of Medicine of the University of Southern California, Los Angeles.

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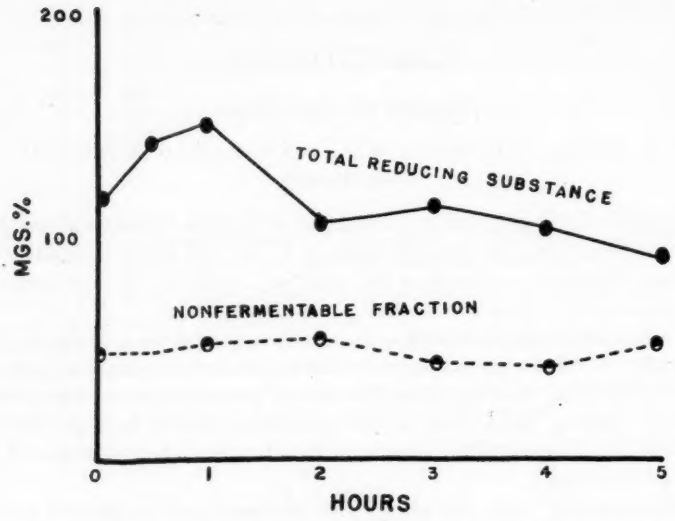
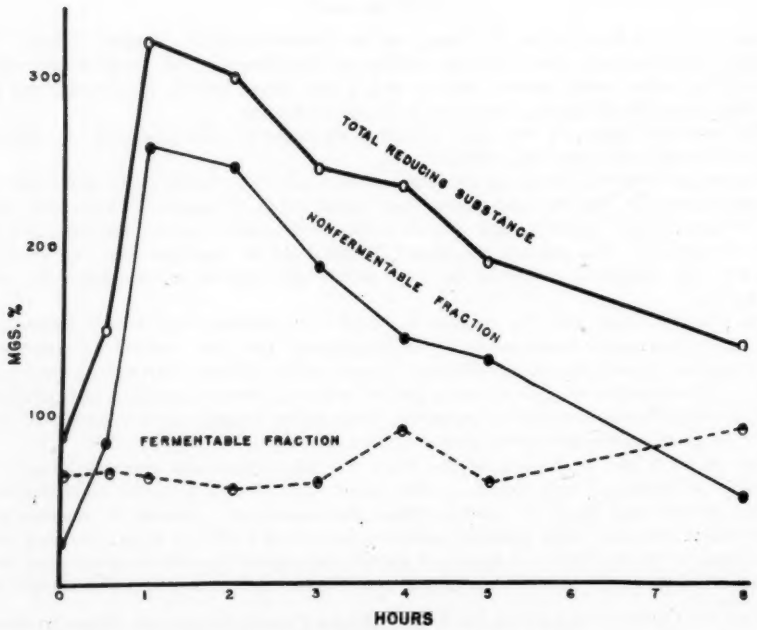


CASE I GLUCOSE TOLERANCE (1.75 GM./KG.)

CHART 1



CASE I GALACTOSE TOLERANCE (1.75 GM./KG.)

CHART 2

Within 10 days, the vomiting subsided and the infant began to gain weight slowly. The urine became sterile. At discharge from the hospital, 4 wk. after admission, she weighed 2500 gm.

When seen in the outpatient department 10 days after discharge, the liver was palpated 4 cm. below the costal margin. To provide a low fat intake, the formula was changed to half skimmed milk. During the next month she did fairly well, gaining weight slowly. Severe vomiting then recurred suddenly and the infant was readmitted to the hospital.

Second Admission (2½ mo. of age): Examination revealed a poorly nourished infant. No evidence of infection or jaundice was found. The abdomen was moderately enlarged, the liver being palpable 4 cm. below the right costal margin. Hgb. was 9.5 gm./100 cc., WBC 7.85 thousand/cmm. with 63% polymorphonuclear leukocytes and 37% lymphocytes. Urinalysis showed 4+ albuminuria and 3+ reduction with Benedict's solution. The icteric index was 6 u. and the NPN 38 mg./100 cc. The total serum protein was 5.0 gm./100 cc. with 3.1 gm./100 cc. albumin and 1.7 gm./100 cc. globulin.

Throughout the second hospitalization of 17 days, the infant did not vomit and remained afebrile. Feedings consisted of an evaporated milk formula. The urinary tract was again thoroughly investigated because numerous urine specimens revealed albuminuria and mellituria. Intravenous pyelogram and retrograde studies were within normal limits. She was discharged after 16 days.

At home she gained slowly and did not vomit, but numerous urinalyses showed constant albuminuria and glycosuria. A diagnosis of galactosemia, previously entertained but discarded, was then reconsidered. When she was 5 mo. of age, the reducing substance in the urine was identified as galactose: (1) by the formation of mucic acid crystals which were identified by the microscopic appearance and melting point (217-218° C. with decomposition); (2) no fermentation with yeast after 3 hr. at room temperature; (3) a positive phloroglucinol reaction.

Third Admission (5 mo. of age): There was a history of low grade fever and vomiting of 1 wk.' duration. Lamellar cataracts were noted for the first time. Because she remained febrile and in such poor condition, the feeding was changed to nutramigen® after 5 days. She improved rapidly in appearance and became afebrile. The galactosuria and albuminuria cleared in 3 and 7 days, respectively. A glucose tolerance test was then performed which was normal (chart 1). The galactose tolerance test showed marked retention of galactose in the blood (chart 2).

The infant was discharged to the outpatient department on a lactose-free diet. During the next 8 mo., she did remarkably well. Weight gain was rapid and steady, averaging 0.5 kg./mo. The most striking change during this period was in the personality. Whereas she had been irritable and cranky, she now became happy, playful and active. The motor development, which had lagged far behind the normal, quickly improved so that she soon compared favorably with an average infant of her age. For the 1st 2 mo. on a lactose-free diet, the liver decreased in size but thereafter remained stationary at 3 cm. below the costal margin. The urine has been consistently free of albumin and galactose. The lamellar cataracts showed no change.

Case 2 (Figs. 1 and 2): T. A. was a male infant of 4 wk. when first seen at the Los Angeles Childrens Hospital. The mother had had 3 previous pregnancies, first a spontaneous abortion and then 2 full term infants (Cases 3 and 4) delivered by cesarian section; both died in this hospital of "liver disease" during the neonatal period. The present pregnancy was complicated by epidemic parotitis 2 wk. prior to term. Delivery was by cesarian section. The patient weighed 2955 gm. at birth and was considered normal until the 6th day of life when fever and marked jaundice developed. During the next 2 wk., he was treated presumptively with penicillin and streptomycin as a neonatal sepsis. In the course of therapy ascites and peripheral edema developed. He improved slowly and was sent home from the maternity hospital at the age of 3 wk. At that time icterus had disappeared but his abdomen was still protuberant. During the week after discharge, the abdomen enlarged and shortly before admission breathing became labored; later the lower extremities became swollen.

Examination on entry revealed an irritable, chronically ill infant. There was 2+ edema of the abdominal wall. The right pupil was oval in shape and eccentrically placed. The sclerae were mildly icteric. Heart and lungs were normal. The abdomen was protuberant and tense and the superficial veins prominent. The liver was palpable 5 cm. below the right costal margin. A few spider hemangiomata were described "upon the skin."

Admission laboratory work included Hgb. 10.5 gm./100 cc., WBC 10.6 thousand/cmm. with 51% polymorphonuclear leukocytes and 49% lymphocytes. The urine was negative except for a trace of reducing substance.

The most probable diagnosis at that time was considered to be cirrhosis of the liver, secondary to infection. Penicillin and sulfadiazine therapy was instituted. On the third hospital day an abdominal paracentesis yielded 150 cc. of clear fluid which was sterile on culture. Two days later he appeared in shock and was ashen gray in color. Although liquids had been accepted well by mouth, he lost large amounts of fluid from the paracentesis wound. The CO_2 combining power was 13 mEq./l. With supportive therapy of blood, plasma and $\frac{1}{6}$ M sodium lactate, improvement was gradual

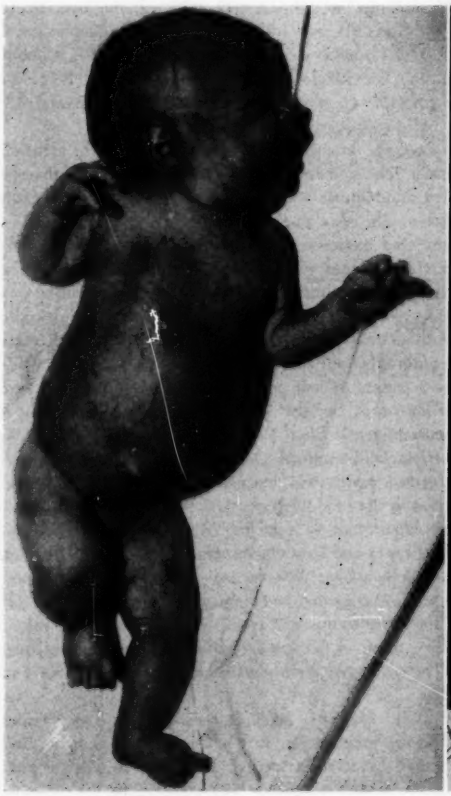


FIG. 1. Case 2, aged 2 mo.

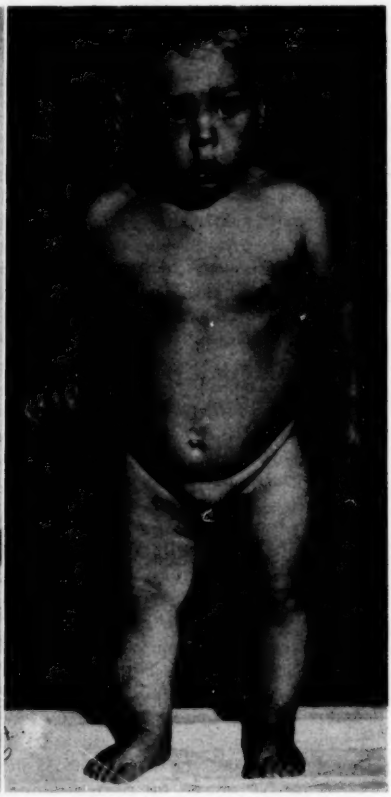


FIG. 2. Case 2, aged 19 mo.

and he was discharged after a hospitalization of 6 wk. At that time the liver edge had receded from 5 cm. to 2 cm. below the costal margin.*

For the 1st 2 wk. in the hospital, all urine specimens revealed albuminuria and glycosuria. Thereafter, the urine was negative except for a trace of albumin.† On entry, blood and urine cultures showed no growth. The total serum proteins were 3.7 gm./100 cc. with a serum albumin of 1.0 gm./100 cc. and a serum globulin of 2.7 gm./100 cc. In 4 wk. these values rose to a total serum proteins of 5.6 gm./100 cc., serum albumin 3.9 gm./100 cc. and serum globulin 1.7 gm./100 cc. The Kahn test was negative, a cephalin flocculation 1+, icteric index 13 u., blood sugar 94 mg./100 cc.

* This may be explained by the fact that soon after entry, because of the enlarged liver, the formula was changed from milk to nutramigen® (lactose-free).

† Again, this is related to the change in formula.

and cholesterol 114 mg./100 cc. The original NPN of 73 mg./100 cc. decreased to 32 mg./100 cc. by the time of discharge.

Second Admission (2½ mo. of age): After 3 days at home the infant was readmitted because he would not move his right arm. The abdomen was still protuberant and the liver was palpable 2 cm. below the costal margin. There was a hard, nonmovable mass at the proximal end of the right humerus. This was aspirated and a few cc. of thick pus obtained; incision and drainage was performed; culture yielded B. aerogenes.

RGs of the extremities revealed a destructive process at the upper end of the right humerus and at the proximal portion of the right femur. Therapy consisted of penicillin, sulfadiazine and strepto-

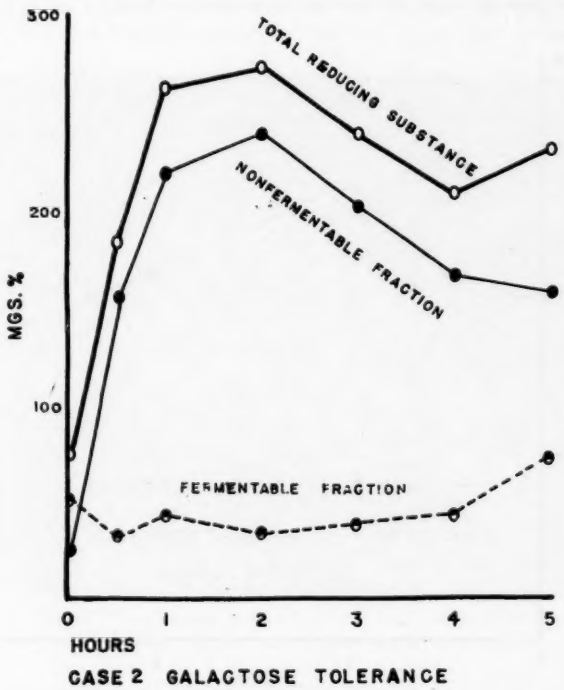


CHART 3

mycin. The hospital course was uneventful and the osteomyelitis healed satisfactorily. During the entire period he was continued on nutramigen® feedings and repeated urinalyses were clear except for an occasional trace of albumin. On discharge after 6 wk., he was much improved. The liver had receded to the costal margin.

On his first visit to the outpatient department 2 wk. later, the formula was changed from nutramigen® to evaporated milk. The liver was still at the costal margin. One month after this return to cow's milk, he reappeared because the abdomen was enlarging again. The liver was palpated 5 cm. below the right costal margin and the spleen was enlarged. During the next 7 mo., the liver edge descended to 7 cm. and urine specimens on consecutive clinic visits showed marked albuminuria and marked glycosuria. Despite this, the infant continued to gain slowly but motor development was definitely retarded.

At 13 mo. of age, the urinary reducing substance was identified as galactose. He was then readmitted for study.

Third Admission (14½ mo. of age): The positive physical findings at this time were early cataracts in both eyes, most marked on the right, and an enlarged liver to 7 cm. below the costal margin. Significant laboratory findings included marked albuminuria and glycosuria, a cephalin flocculation of 1+, an icteric index of 18 u., NPN of 37 mg./100 cc. and a prothrombin time of 23.5 sec. with a control of 12.5 sec. Glucose and galactose tolerance curves were done (charts 3 and 4). Galactose tolerance curves done on the parents were normal.

He was discharged on a lactose-free diet, nutramigen®. During the next 7 mo., with repeated visits to the outpatient department, he gained about 0.6 kg./mo. Coincidently, the mental and physical development improved rapidly, so that at 19 mo. he was running about actively and beginning to talk. The liver had decreased in size to 3 cm. below the costal margin and urinalyses had been consistently negative. However to date, the cataracts have persisted unchanged.

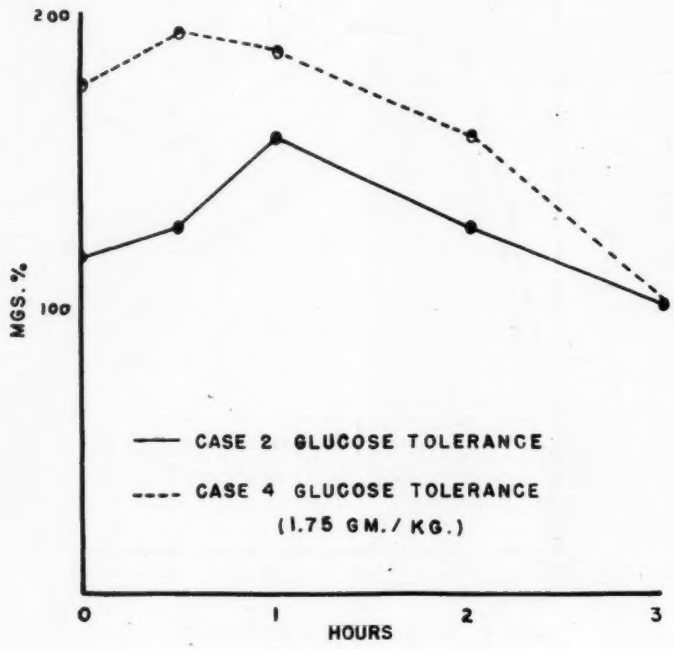


CHART 4

Case 3. G. A. (sibling of Case 2), a 4 wk. old white male, was admitted to the Los Angeles Childrens Hospital with jaundice, vomiting and diarrhea of 3 wk.' duration. He was the first live child of healthy parents. The delivery was by cesarean section because of a difficult breech presentation. The infant was apparently normal until the sixth day of life when he became noticeably jaundiced. This was soon followed by vomiting and mild diarrhea. The only treatment consisted of gavage feedings of breast milk. The vomiting became more severe and the day before admission was bile-stained.

Examination revealed a moderately ill infant weighing 2727 gm. Temperature was normal and the respiratory rate was 40/min. The left tympanic membrane was inflamed. The lungs and heart were normal; the liver was palpated 4 cm. below the right costal margin.

Admission laboratory data included Hgb. 11.5 gm./100 cc., RBC 3.99 million/cmm., and WBC 8.4 thousand/cmm. with 44% polymorphonuclear leukocytes and 56% mononuclears. The urine revealed 4+ albumin and 2+ glycosuria with no cellular elements or acetone.

The infant was believed to have a sepsis and was treated with sulfathiazole and parenteral fluids. The vomiting and diarrhea soon subsided and breast milk was accepted by bottle. Five days after admission a blood culture grew hemolytic Staph. aureus, coagulase positive. Because of the persistent albuminuria and glycosuria, a fasting blood sugar was drawn which was 289 mg./100 cc. On the basis of hyperglycemia and the marked urinary reducing substance, small doses of protamine zinc insulin were tried with no decrease in urinary reducing substance. On the 14th hospital day the temperature rose sharply and became septic in type. In the next 8 days, the course was gradually downhill and he died. An autopsy was performed.

Autopsy Findings

Gross Findings (Dr. Ralph E. Knutti): The body was that of a markedly emaciated white male infant. The anterior fontanel was depressed. There was dry subcutaneous tissue with no evi-

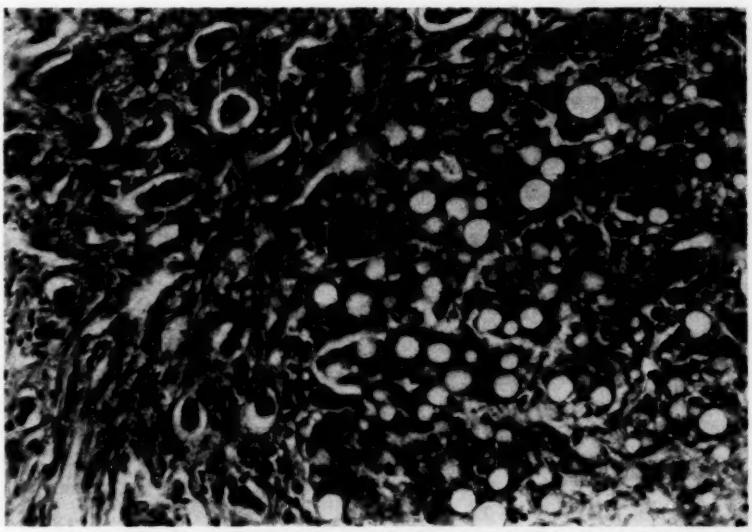


Fig. 3. Case 3. Section of liver showing periportal fibrosis with increased number of bile ducts. Large clear vacuoles represent intracellular deposits of neutral fat.

dent fat. The muscles were pale and poorly developed. Grossly, all organs appeared normal with the exception of the liver and pancreas which will be described.

Liver (see Fig. 3): The liver weighed 200 gm. (normal 133 gm). It was enlarged and had a yellowish-orange color. The capsule was dull and of a pigskin texture. On section, the surfaces were a buff color and had a roughened appearance caused by alternating slightly raised and slightly depressed areas. The gall bladder contained a small amount of yellowish-green, rather thin bile. The extrahepatic bile ducts were patent.

Microscopically the liver showed marked periportal fibrosis. The fibrous tissue was dense and infiltrated the margins of the lobules with strands extending into them. There was atrophy of the liver cells immediately adjacent to the connective tissue. Elsewhere, all the liver cells were filled with large, pale, fat-containing vacuoles. There were numerous bile ducts in the biliary areas. The bile canuliculae were greatly dilated and filled with brown pigment. Occasionally, some polymorphonuclear leukocytes could be seen about the small bile ducts. Sections through the cystic and common ducts showed these structures to be patent, and there was no evidence of acute or chronic inflammation.

Pancreas: The pancreas was swollen and red in color. This was most prominent on the anterior superior surface near the tail. On cut section there were bright red areas between the lobules.

TABLE 1

Authors		Age	Sex	Positive Family History	Abnormal Development		Physical Findings					Laboratory Findings								On Milk-Free Diet			
					Mental	Physical	Jaundice	Cataracts	Hepatomegaly	Splenomegaly	Ascites	Albuminuria	Glycosuria	Ketonuria	Urinary Galactose Identified	Abnormal Galactose Tolerance	Abnormal Glucose Tolerance	Abnormal Liver Function	Hgb. (gm.)	Cataracts Improved	Hepatomegaly Decreased	Albuminuria Disappeared	Glycosuria Disappeared
Von Reuss[1]	1908	8 mo.				+			+	+		+	+	−	+						+		
Bossert[13]*	1916	inf.	M			+	+	+	+	+	−	+	+	−					11.2				
Goppert[2]	1917	2½ yr.	M	+	+	+	+		+	+		+	+	−	+	+					+	+	+
Boer[26]* (1)	1932	7 yr.	F	+		+			+				+	−									
(2)*		1 yr.	F	+		+			+				+	+		+							
Unshelm[3]*	1934	3 mo.	F	−		+			+	+			+	−		+	−				+		+
Mason and Turner[4]	1935	6 mo.	M	−		+	−	+	+	+			+			+	−		9.4		+	+	+
Norman and Fashena[5]	1943	11 wk.	M	−	−	+			+	−		+	+	−	+	+	−	±	7.9		+	+	+
Mellinkoff[6]	1945	2 mo.	M	−		+	+		+		+	+	+	−	+		−	+	11.5		+	+	+
Bruck and Rapoport[7]	1945	9 wk.	M		+	+	+	+	+	+		+	+	−	+	+			7.7	+	+	+	+
Dunsky[14]*	1946	2 mo.	M	−		+	+		+	−	+	+	+	−				+					
Goldbloom and Brickman[8]																							
(1)	1946	6 mo.	M		+	+	+	+	+	+		+	+	−	+	+	±		14.3	sl.	+	+	+
(2)		3½ mo.	F			+	−	+	+			+	+	−	+	+			8.0	sl.	+	+	+
Goldstein and Ennis[9]	1948	2½ mo.	M	−		+	+	+	+			+	+	−	+	+	−	±	8.0	+	+	+	+
Greenman and Rathbun[10]	1948	18 wk.	M			+	+	+	+	+	+	+	+	−	+	+	−	−		+	+	+	+
Bell[11]* (1)	1950	1 wk.	F	+			+		+										16.0				
(2)		1 wk.	M	+		+	+	−	+	+		+	+	−	+	+	−	−	17.0		+	+	+
Donnell and Lann																							
(1)	1950	5½ mo.	F	−	+	+	+	+	+	+		+	+	−	+	+	−	−	9.5	−	+	+	+
(2)		13 mo.	M	+	+	+	+	+	+	+	+	+	+	−	+	+	−	±	10.5	−	+	+	+
(3)*		4 wk.	M	+		+	+		+			+	+	−					11.5				
(4)*		5 wk.	F	+		+	+		+	+	+	+	+	−			−		8.7				

* Clinical picture typical of galactosemia but glycosuria not identified.

Explanation of symbols:

\+ Stated by author to be present.

− Stated by author to be absent.

Blanks indicate no statement by author.

Microscopically there were areas of hemorrhage into the interlobular tissue with occasional focal hemorrhage in the lobules. Numerous foci of polymorphonuclear leukocytes were present. The pancreatic acinar cells adjacent to such areas were necrotic. The tissue elsewhere was normal.

Anatomic Diagnosis: 1. Cirrhosis of the liver; 2. Acute hemorrhagic pancreatitis.

Case 4. Th. A. (sibling of Cases 2 and 3), a 5 wk. old white female, was admitted to the Los Angeles Children's Hospital. She was the second living child of healthy parents. Delivery was by cesarean section after an uneventful gestational period. Birth weight was 3270 gm. At the age of 5 days she became jaundiced and was treated with penicillin and sulfathiazole for 1 mo. at another hospital. The jaundice did not clear, however, and the infant was transferred to this hospital.

Examination on admission revealed a fairly well nourished infant in no acute distress. The skin and sclerae were definitely icteric. The abdomen was protuberant and the liver was palpable at the level of the umbilicus. The spleen was felt 2 cm. below the left costal margin.

On admission, the Hgb. was 8.7 gm./100 cc. and the WBC 26.3 thousand/cmm. with a normal differential. The urine revealed 4+ albumin and 3+ glycosuria without ketonuria or cellular elements in the sediment. A blood culture drawn on entry was negative. All urinalyses carried out during hospitalization showed marked albuminuria and glycosuria. Because of the persistent glycosuria, a glucose tolerance test, performed during the third week in the hospital, showed a high fasting blood sugar but an essentially normal tolerance curve (see chart 4).

Therapy started on admission included penicillin and sulfathiazole. Feedings consisted of a lactic acid milk formula. The infant's abdomen became more distended and ascites became clinically evident. On the seventh hospital day, an abdominal paracentesis yielded 125 cc. of clear fluid which was sterile on culture. The following week 5 additional paracenteses totaled 560 cc. in volume. Afebrile during the 1st 5 wk. of hospitalization, she suddenly developed a fever of 39.4°C.; in the next 2 days she went rapidly downhill and died.

Permission for autopsy was not granted.

Discussion

The importance of identifying the specific reducing substance in patients showing glycosuria can not be overemphasized. Early diagnosis and treatment will often result in rapid clinical improvement. The significant findings in the cases reported in the literature to date appears in table 1. The outstanding features of galactosemia are failure to gain weight, jaundice, hepatomegaly, albuminuria, galactosuria, lamellar cataracts and the characteristic improvement on a lactose-free diet. Cataracts are reported in 8 of the 21 cases; on a galactose-free diet they disappeared in three, improved in two, remained unchanged in two and the outcome is unknown in one.* In Cases 1 and 2 reported here, an ophthalmologist has not been able to note any significant changes in the appearance of the cataracts.

The appearance of jaundice in the neonatal period and its persistence for a variable length of time is a fairly constant feature. Abnormal liver function tests are an unusual finding. That ketonuria has never been demonstrated in galactosemia is most unusual, since the patients are in a relative state of starvation.

Cases 1 and 2 fit the criteria established for the diagnosis of galactose diabetes. The ascites, edema and focal abscesses noted in Case 2 were an unusual finding that simulates the case reported by Mellinkoff et al.[6] No mention of serum albumin concentrations was made in their paper although the total serum proteins were low. On a lactose-free diet, the serum proteins in their patient rose rapidly and the ascites and edema disappeared simultaneously. In Case 2, at the height of the edema, the serum proteins were 3.7 gm./100 cc. with a serum albumin of 1.0 gm./100 cc. Because of vomiting the infant was placed on nutramigen® feedings and the edema and ascites gradually disappeared.

* Bruck and Rapoport[7] report the presence of cataracts in Mason's patient, but the end result is not known.

After one month the proteins had risen to 5.6 gm./100 cc. with a serum albumin of 3.4 gm./100 cc. Greenman,[10] on the other hand, reported a case with ascites with normal proteins. In cases reported without edema, hypoproteinemia with a low serum albumin was not found. The ascites, edema and venous distension is thought to be the result of portal hypertension.[12] However, the presence of this complication in the patients with hypoproteinemia and the absence in those with normal serum proteins would suggest that this is an additional, if not the principal, cause.

In Cases 3 and 4, two siblings of Case 2, galactosemia was not suspected before death. In retrospect, the persistent albuminuria and glycosuria, in addition to the clinical picture, should have suggested the diagnosis. Infantile diabetes mellitus was considered in Case 3 and insulin was given as a therapeutic test. No effect on the glycosuria after three daily injections suggests that the child was excreting a sugar other than glucose. In Case 4, ascites and edema were found in addition to the usual signs and symptoms. Although the fasting blood sugar was high in this infant, the oral administration of glucose did not result in an abnormal tolerance curve, which suggests either a normal utilization or a decreased absorption of glucose. The high fasting blood sugar could be explained by the presence of an additional reducing substance, probably galactose.

Until recently, little has been reported concerning the pathology of the liver in this disease. Von Reuss[1] demonstrated at necropsy a typical cirrhosis in an infant with galactosuria and hepatomegaly of the Laennec type. Two additional reports,[13, 14] including autopsy studies, have been found in the literature in which the diagnosis was not definitely established.

Bell et al.,[12] in 1950, contributed to our knowledge of liver pathology with biopsy studies on a proved case of galactosemia. The biopsy revealed a fatty liver with early fibrotic changes. Bell also reported findings at necropsy on the sibling in whom the disease was suspected. The enlarged liver showed severe, diffuse fatty infiltration of most of the parenchymal cells but no cirrhosis. The finding of a fatty, cirrhotic liver at autopsy in the present Case 3 is in accord, then, with Bell's observations. It is difficult to evaluate the part played by the hemorrhagic pancreatitis in Case 3. It is felt, however, that this probably represented a terminal event.

In 1935 Mason and Turner[4] first proposed an hypothesis to explain the etiology of galactosemia. They felt that the primary difficulty was due to a failure in conversion of galactose into glycogen. They demonstrated that a rise in the blood galactose concentration caused depression of the blood glucose level, and concluded that a relative glucose starvation existed; presumably the clinical picture resulted from this process. Bruck and Rapoport,[7] on the other hand, suggested that galactose itself exerted a toxic action. They found that the blood glucose depression was not a constant finding. Norman and Fashena[5] presented data which substantiated Mason's hypothesis.

The autopsy findings in Case 3 are in agreement with Bell's hypothesis that fatty infiltration may play a part in the etiology of hepatomegaly in galactosemia. Such a mechanism might account for the rapid fluctuations in liver size that have been observed.[7] Fat may be deposited and removed from the liver very rapidly in the experimental animal.[15] That congestion and edema of the liver or storage of some substance other than fat may be a factor in the liver enlargement cannot be ruled out on the present available data.

The etiology of cirrhosis in these patients has not been satisfactorily explained. It is accepted that fatty infiltration of the liver may precede or accompany cirrhosis, but it is also well appreciated that cirrhosis does not invariably follow fatty changes of that organ.

Whether the fatty infiltration or portal cirrhosis is due to direct toxic action of galactose on the liver or to relative glucose starvation or possibly to both, is difficult to evaluate. It is known that many hepatotoxins such as chloroform, carbon tetrachloride and phosphorus interfere with liver metabolism so that fat accumulates followed by degeneration of liver cells.[16] In the experimental animal, a high galactose intake produces serious toxic manifestations but direct injurious effect on the liver has not been described.[17] Starvation states may also produce a fatty liver.

Worthy of comment is that ketosis is not reported in the proved cases of galactosemia while it is common in other diseases associated with fatty cirrhotic livers. Bell's report of normal liver glycogen content in a biopsy specimen from a patient with galactosemia is not out of accord with fat deposition in the liver since normal or even abnormal deposits of glycogen can accompany fatty changes in that organ.[18] That these glycogen stores can readily be mobilized has been demonstrated on numerous occasions. It is doubtful, however, that this plays a part in the prevention of ketosis. It has been stated that substances which damage the liver decrease ketosis.[19] The toxic effect of galactose on the liver might thus be a factor in the prevention of ketosis.

The discussion so far has dealt with the effects of the high concentration of galactose in the blood of these patients. The primary defect is not yet known. Stimulated by excellent work on the red bread mold, Neurospora crassa, attention has been focused on genetics to explain the etiology of obscure metabolic disorders.[20–22] It was shown that single genes controlled individual biochemical reactions and that when mutation occurred, the specificity of the gene was altered; in this fashion an individual lost the ability to carry out a specific biochemical reaction. Conidiae of this mold were treated with ultra violet light or x-ray which is known to increase mutation rates.[23] Mutant strains were obtained which differed from the parent strain in that growth could be accomplished only on complete media, whereas the parent mold would grow on simpler media. For example, mutant strains were obtained which required the presence of vitamins B_1, B_2 and B_6 in the media to promote growth, while the parent strain did not require these substances.[23] This relationship between heredity and chemical reactions has been appreciated for many years.[24] Alkaptonuria, an inherited condition, characterized by lack of an enzyme necessary for the metabolism of tyrosine, is a good example. In 1950, in a review of *Inborn Errors of Metabolism,* Taggart and Mason have included galactosemia in this group of diseases.[25]

That galactosemia is genetically determined is strongly suggested by a number of reports. Very early Goppert intimated this in the report of a patient with four siblings who had liver disease.[2] Boer[26] also described two siblings with retarded development, hepatomegaly and the presence of a reducing substance in the urine. Galactose intolerance in both of his patients was demonstrated by oral galactose tolerance curves. He received information that a third sibling, not under his care, had died in infancy of an identical condition. In 1950, Bell et al. also reported the disease in siblings.[12] The present report of the syndrome in three members of one family further emphasizes the hereditary aspects.

In combining all the features of this discussion, the possibility exists that a specific gene necessary for the metabolism of galactose is absent, or more likely, altered so that the utilization of this sugar is partially or completely impaired. This results in an accumulation of galactose in the blood stream which, in turn, exerts a direct toxic effect upon certain tissues. The cataracts and the fatty infiltration of the liver are the most important manifestations of this toxicity; mental retardation and renal disturbances are probably additional evidence.

SUMMARY

Four cases of galactosemia are presented; three occurred in one family. Autopsy findings on one case revealed portal cirrhosis and fatty infiltration of the liver.

The etiology of hepatomegaly and the genetic aspects of the disease are discussed. The importance of identifying the reducing substance in patients with glycosuria is emphasized by the cases reported here.

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SPANISH ABSTRACT

Galactosemia; Reporte de Cuatro Casos

Se describen cuatro casos de galactosemia, tres de los cuales ocurrieron en una misma familia. El estudio necropsico en un caso demostró una cirrosis portal y infiltración grasosa del higado.

Los sintómas clinicos comunes a todos los pacientes fueron: un retardo del crecimiento y desarrollo normal, vómitos, ictericia y hepatomegalia. En dos casos se observaron cataratas lenticulares. Albuminuría y glicosuría fueron signos constantes en todos los casos. Estudios del metabolismo carbohidrato en dos pacientes reveló una intolerancia marcada a la galactosa. Estos pacientes obtuvieron una mejoria clinica con la eliminación de la lactosa de la dieta.

Los autores discuten la etiología de la hepatomegalia y los aspectos genéticos de la enfermedad.

Se indica la importancia en la identificación de substancias reductoras en pacientes con glicosuría en el diagnóstico de la galactosemia.

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HYPERPOTASSEMIA AND BODY WATER DISTRIBUTION IN AN ANURIC CHILD

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CONSERVATIVE management of anuria has in many instances prolonged life.[1, 2] This involves the correction of water and electrolyte deficits and the utilization of the protein-sparing effects of glucose to decrease the rate of tissue catabolism. These measures attempt to preserve the integrity of the body fluids, thus retarding the accumulation of toxic substances, i.e., potassium, and reducing the risk of water intoxication and possible pulmonary edema which often results from vigorous fluid, water administration in anuric patients.

Attempts to simulate renal excretion by peritoneal lavage,[3-6] gastrointestinal irrigation,[7-9] exsanguino-transfusion[10, 11] and the artificial kidney technic[12-14] seek to maintain homeostasis until there is a return of renal function. Therapy directed at restoration to normal of body water and electrolyte requires knowledge of the normal distribution and of the factors influencing shifts of these substances between the extra- and intracellular compartments.

The following is a report of clinical observations and studies of body water distribution during the course of anuria with progressive hyperpotassemia in a 6 year old child.

CASE HISTORY

L. T., a 6 yr. old Negro female, was admitted to the Children's Medical Service, Bellevue Hospital, after anuria of 4 days' duration and vomiting and constipation of 6 days' duration.

Family history was not remarkable other than that the mother had been treated for syphilis 13 yr. previously. There was no known renal disease. Past history revealed that gestation and delivery were uncomplicated except for a repeat course of antiluetic therapy for the mother. Neonatal course, growth and development were normal. Immunizatiops included diphtheria, tetanus, pertussis and smallpox vaccination. Previous illnesses included rubella, rubeola, epidemic parotitis, and varicella. Review of systems revealed frequent colds, 2 to 3 each winter lasting 2 to 3 wk., and occasional chronic cough with slight production of yellow sputum, but no known streptococcal infection. There were no previous genitourinary tract difficulties, hematuria, pyuria or oliguria prior to the present illness.

The patient had been well until 2 wk. prior to admission when she began to cough. Eight days prior to admission the onset of anorexia, malaise and fever occurred. The family physician diagnosed tonsillitis and cervical adenitis and prescribed rhubarb and soda and aspirin. Six days prior to admission, fever and anorexia continued, vomiting after meals and constipation were noted, and the child was more somnolent than usual. She urinated normally 7 days prior to admission.

Five days prior to admission she continued febrile and was examined at another hospital where sulfadiazine was prescribed. The amount ingested by the patient is not known, but it is thought to be small because the medication was vomited more often than retained. Symptoms continued as before. Three days prior to admission, the patient was said to have passed a small amount of smoky urine

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which was not seen by the mother. This is the last known voided specimen. Vomiting, headache, periumbilical pain, malaise and anorexia continued until admission.

Anuria was essentially of 7 days' duration, except for the possible small specimen 3 days prior to admission. No history could be elicited of ingestion of toxic substances other than the few doses of sulfadiazine.

Physical examination revealed a well-developed, well-nourished alert cooperative 6 yr. old female Negro child who did not appear acutely ill. The rectal temperature was 36.7° C., respiratory rate 26/min., pulse rate 68/min., blood pressure 125/88 mm.Hg and wt. 26.5 kg. The breath had a uremic odor, but the face was not puffy. The pharynx was minimally injected. The lungs were clear to percussion and auscultation and the heart sounds were of good quality. A soft medium-pitched blowing systolic murmur was heard over the entire precordium, maximal at the fourth intercostal space at the left sternal border. There was moderate costovertebral angle tenderness bilaterally but no abdominal masses or peripheral edema were observed. Otherwise the physical examination was normal.

Hgb. 11 gm./100 cc. (Sahli), RBC 4.3 million/cmm., and WBC 15.5 thousand/cmm. with a

TABLE 1

BLOOD CHEMICAL ANALYSES*

Date	Time	Blood NPN mg./100 cc.	Serum CO_2 Combining Power mEq./l.	Serum Cl mEq./l.	Serum Na mEq./l.	Plasma K mEq./l.	Blood Sulfadiazine mg./100 cc.	Serum Ca mg./100 cc.	Serum P mg./100 cc.	Serum T.P. gm./100 cc.	A/G	Body Wt. kg.
6/10	10 a.m.	193	12.5	111	133	7.0	0.89			8.57	4.62 / 3.95	26.5
6/11	10 a.m.		12.5	87	129	7.0						26.5
6/11	6 p.m.			91	140	8.0			5.7	5.6		
6/12	6 a.m.			90	139	9.4						
6/12	5 p.m.			76	130	8.2						25.9
6/13	10 a.m.	292	13.8	85	139	9.5				6.2		27.0
6/14	4 a.m.	284		79	130	10.1		9.95	10.4	4.9		

* From the chemistry laboratory of The Children's Medical Service.

normal differential. Catheterization of the bladder yielded a few cc. of yellow urine, pH 5.5, albumin negative. The sediment contained a few white blood cells but no crystals or other formed elements. Initial blood chemical analyses showed: blood NPN 193 mg./100 cc., blood sulfadiazine concentration 0.89 mg./100 cc., serum CO_2 combining power 12.5 mEq./l., serum sodium 133 mEq./l., plasma potassium 7 mEq./l. (table 1).

The patient remained afebrile and in good general condition over the 1st 24 hr. Occasional vomiting persisted. In view of the history of sulfadiazine ingestion and the finding of sulfonamide in the blood, bilateral ureteral alkaline washouts were done under light ether anesthesia. No urine was obtained from either ureter. The irrigation fluid revealed no sulfonamide crystals nor could any be detected by chemical analysis. Ureteral catheters remained in situ for 18 hr., after which they were removed and a Foley catheter placed in the bladder. During the 1st 24 hr. minimal intravenous fluids were given (300 cc. of 5% glucose and 50 cc. of 5% sodium bicarbonate). The electrocardiogram revealed no definite changes indicative of hyperpotassemia (Fig. 1A).

The child continued alert on the second hospital day. Blood pressure and pulse remained normal. The chest was clear and there was no evidence of edema. Studies started 42 hr. after admission are described below. Fluid intake totaled 275 cc. (110 cc. normal saline, 50 cc. 10% inulin, 50 cc. 25% mannitol, 34.5 cc. 99.8% deuterium oxide and 30 cc. water). Feedings of glucose solution were vomited.

Her condition appeared to be good on the third hospital day. Fluids tolerated by mouth included 470 cc. orange juice, 120 cc. water and 70 cc. of 5% sodium bicarbonate. No intravenous fluids were given. Blood pressure was normal. Tissue turgor was good although muscle tone was poor. A fluid wave in the abdomen was noted. Weight remained stationary.

On the fourth hospital day the blood pressure was normal, respirations were slightly labored, but the lungs remained clear. The patient was more lethargic. Slight puffiness about the eyes and definite pitting edema of the lower extremities were evident. Skin turgor was fair. The blood NPN concentration was markedly elevated and the plasma potassium concentrations of blood samples drawn on previous days were observed to be at subtoxic levels (table 1). Fluid intake was limited to 610 cc. of orange juice with added sodium bicarbonate. The ECG now showed minimal evidence of potas-

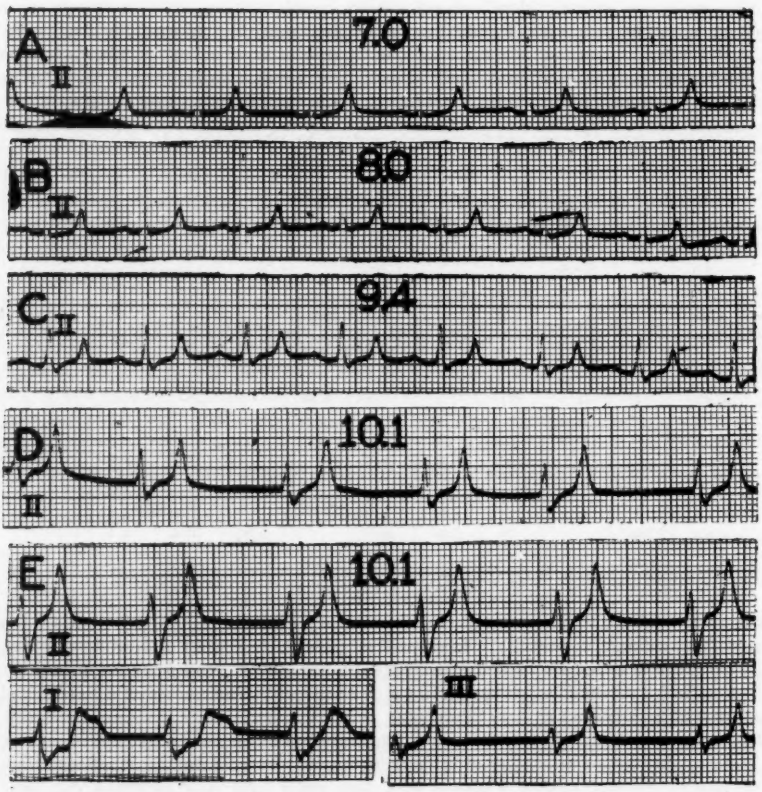

FIG. 1. Electrocardiographic tracings and plasma concentrations of potassium in mEq./l. on last 5 days of anuria. Standard lead II is shown in all tracings; in E, final tracing, leads I and II are also shown.

sium intoxication, widening of the QRS complex, depression of the ST segment, elevated T waves and relative shortening of the Q-T interval (Fig. 1C). At 10:55 p.m., the patient was noted to have a slow irregular pulse at a rate of 50/min. ECG revealed marked potassium intoxication as evidenced by the ventricular rhythm with widened QRS, apparent auricular paralysis and the appearance of an even higher T wave (Fig. 1D). Ten cubic centimeters of 5% calcium gluconate were given intravenously with a transient increase in the heart rate and a return to a regular rhythm.

It was considered desirable to attempt to reduce the plasma potassium concentration by the technic of exsanguino-transfusion[10, 11] since no other means of effecting dialysis was available. After a biologic test transfusion for compatibility with 50 cc. of blood, the procedure was started. However, the ECG revealed extreme potassium intoxication (Fig. 1E) and the patient died about 1 hr. after the

exsanguino-transfusion was started. The patient was alert and cooperative until the last few hours. No autopsy was performed. The clinical diagnosis was acute renal failure, etiology unknown.

Methods of Study

Total body water was measured by the in vivo dilution of deuterium oxide (heavy water). The volume of extracellular water was measured by the in vivo dilution of inulin. The volume of intracellular water was then obtained by subtracting the inulin volume of distribution from the deuterium oxide volume of distribution. Plasma volume was measured with T 1824.

On the second hospital day, 101.7 cc. of a solution of 5% inulin and 12.5% mannitol were injected intravenously with a constant infusion at a rate of 3.39 cc./min. This was followed 9 hr. later by an intravenous injection of 34.5 cc. of deuterium oxide from a calibrated syringe. Before the injection, a control blood sample was obtained for determination of the plasma blank concentration,

TABLE 2

Body Water Distribution

Experimentally obtained values for distribution of body water on 2 days during course of anuria are here compared with accepted values for normal water distribution.

Accepted Normal Values	Total Body Water	% of Body Weight	Extracellular Water	% of Body Weight	Intracellular Water	% of Body Weight
		70%		20%		50%

Experimental Values	Deuterium Oxide Space (D_2O)		Inulin Space		Deuterium Oxide Space minus Inulin Space	
	l.	% Body Wt.	l.	% Body Wt.	l.	% Body Wt.
June 12, 1949 6:46 a.m.	18.2	69	9.0	34	9.2	35
June 14, 1949 4:00 a.m.	21.1	78	12.7	47	8.4	31

B_o, for subtraction from subsequent plasma concentrations of inulin and mannitol. The volume of distribution of inulin and mannitol was calculated as the total amount injected (in mg.) divided by the concentration in plasma water (in mg./cc.). The deuterium space was calculated as the amount of deuterium oxide injected divided by the concentration of deuterium oxide in plasma water corrected for the natural occurrence of deuterium (0.02 atoms %).

Samples of blood were drawn at 1 to 12 hr. intervals until the patient's death. On 2 occasions the T 1824 space was measured from a single sample drawn 10 min. after injection and read against a control plasma blank drawn just before the dye was injected.

Inulin was determined by Harrison's modification[15] of the method of Alving, Rubin, and Miller,[16] mannitol by the method of Corcoran and Page[17] and deuterium oxide was measured in the mass spectrometer.*

Equilibrium distribution of inulin was attained in this patient between 4 and 9 hr. after injection. At 6:46 a.m. on June 12, 1949, 14 hr. after the injection of deuterium oxide, simultaneous inulin and deuterium oxide spaces were 9.0 and 18.2 l., respectively, the difference being 9.2 l. (table 2). At 4:00 a.m., June 14, 1949, 45 hr. later and 2 hr. before death, repeat determinations revealed an

* The authors are indebted to Dr. Robert Jailer of the Sloan-Kettering Institute for the deuterium analysis with the mass spectrometer.

inulin space of 12.7 l. and a deuterium oxide space of 21.1 l., the difference being 8.4 l. The latter deuterium oxide space is corrected for an assumed insensible water loss of 30 cc./kg. body weight/day. The mannitol space was 10 l. at 6:46 a.m. on June 12, 1949, and 14.1 l. at 4:00 a.m. June 14, 1949. The T 1824 space was 1,320 cc. at 7:00 p.m. on June 11, 1949, and 1,300 cc. when measured again 7 hr. before death at 8:05 p.m. on June 13, 1949. The hematocrit varied between 25.5 and 29.5 with no consistent trend over the 4 day period of observation. Urine flow was less than 50 cc. throughout the course.

Discussion

Accepting the initial inulin space as a measure of the extracellular water volume, and the initial deuterium oxide space as a measure of the total body water, the preceding data demonstrate an abnormally increased extracellular fluid volume in the presence of a normal total body water after a week of anuria (table 2).

Errors in the deuterium space arise from extrarenal losses of deuterium oxide and exchange of deuterium with available tissue hydrogen. The final deuterium oxide space has been corrected for the estimated insensible water loss.*

Evidence that inulin is distributed in the extracellular space and, under normal circumstances, is not metabolized has been summarized elsewhere.[18-20] The fact that the initial mannitol space was greater than the inulin space probably is due to the metabolism of mannitol rather than to inert penetration of tissue cells.[21-23]

Errors in the inulin space arise from possible cell penetration or metabolism after prolonged retention of inulin in anuria. Berger and associates[24] have evidence suggesting that inulin is metabolized in the nephrectomized dog. However, if these sources of error were insignificant, and the final inulin space measured the extracellular space, one would be forced to postulate osmotic inactivation of cell electrolyte.

For these reasons, the authors hesitate to conclude that in this patient there occurred the indicated final decrease in the intracellular fluid compartment, although the direction of change of water from the intracellular compartment is consistent with (1) tissue destruction (starvation) with release of intracellular water and electrolyte, (2) water of oxidation and (3) osmotic attraction to the extracellular compartment as a result of insensible water loss and administered sodium salts.

Fluid and electrolyte therapy in this case was directed toward maintenance of the body weight and correction of the acidosis by administration of sodium bicarbonate and water.[1,2] Unfortunately, insufficient glucose was administered so that maximal retardation of tissue catabolism was not obtained as suggested by the rapidly increasing azotemia, phosphatemia and hyperpotassemia (table 1). On the two days prior to her death, the patient received 1,080 cc. of orange juice[25] with added sodium bicarbonate and dextrose. It was recognized subsequently that this amount of orange juice constituted an additional burden of 47 mEq. potassium. Thus, in this patient a rapid rise in plasma potassium concentration from 7.0 to 10.1 mEq./l. occurring over a four day period is in part attributable to orally administered K present in orange juice. Peters[26] has emphasized recently that almost all food contains K and should be withheld in the management of the anuric patient.

During the first 48 hours, when the plasma potassium concentration was 7.0 mEq./l., the ECG showed no evidence of potassium intoxication. Widening of the QRS and early elevation of the T wave occurred when the plasma concentration was 9.4 mEq./l.

* The fact that the weight increment was less than the deuterium oxide space increment (table 1) may be attributed to weighing errors and/or underestimation of the insensible water loss.

The onset of the terminal episode consisted of arrhythmia, bradycardia, absent P waves, premature ventricular contractions from varying foci of origin, widening of the QRS complex and marked elevation of the T waves. Ten cubic centimeters of 5% calcium gluconate was given intravenously with a transitory return to a regular rhythm and increase in the heart rate. Saline-lactate solutions in small amounts had no effect on the ECG. Progressive signs of potassium toxicity continued (Fig. 1C) until terminal disorganization of the ventricular complex was noted just preceding cardiac arrest. The continuous electrocardiographic changes and the high plasma potassium concentrations are sufficient evidence that the terminal episode was due to potassium intoxication.

Hoff, Smith, and Winkler[27] have shown in dogs that death following nephrectomy or bilateral ureteral ligation results from potassium intoxication. Previously these authors described the serial changes in the ECG occurring with potassium intoxication in normal animals.[28] They also noted that simultaneous administration of calcium diminished the effects of K on the cardiac conduction system although the usual toxic effects of K could be obtained at a higher plasma concentration of K.[29]

Normal humans rapidly adjust to the intake of potassium salts by increased renal excretion[30-33] and uptake of K by the intracellular compartment.[30, 31] Even though patients with moderate to severe nephritis may excrete sufficient K daily to maintain a normal plasma concentration in the presence of tissue catabolism and moderate potassium intake, the principal limiting factor in the excretion of K probably is a reduction in the glomerular filtration rate.[30, 34, 35] Certainly in anuria there is no other significant route of egress for K except for minimal skin and gastrointestinal losses. An increasing plasma potassium concentration with potassium intoxication is an ever present danger.[36, 37] Finch[38] was able to ameliorate the symptoms of potassium intoxication with intravenous sodium salts; however, both patients succumbed shortly thereafter. He also treated one case with calcium gluconate intravenously with results comparable to those of the present investigators.

Recently Levene et al.[39] described electrocardiographic changes associated with potassium intoxication in 16 cases, most of whom were anuric. They noted that the electrocardiographic manifestations of hyperpotassemia were potentiated by a diminished serum sodium. Reversal of the findings after administration of sodium salts occurred temporarily in a few cases. Many others have reported electrocardiographic changes of hyperpotassemia in patients with renal insufficiency.[40, 41]

Treatment of hyperpotassemia may now be approached by use of an ion-exchange resin or by dialysis with the artificial kidney. With the latter technic, recently several groups have been able to lower the plasma potassium levels significantly.[42-45] By proper adjustment of the dialysing medium this could be accomplished similarly by peritoneal lavage. While exsanguino-transfusion is the only alternative in the absence of facilities for the other methods, it is the least desirable, since it entails the hazards of a transfusion reaction and requires massive exchanges to significantly lower the plasma potassium concentration.

Summary

A patient with anuria in whom a rising plasma potassium concentration was associated with progressive signs of potassium intoxication exhibited a decrease in the intracellular compartment and increase of the extracellular compartment without significant change in plasma volume, while total body water remained essentially normal.

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SPANISH ABSTRACT

Hiperpotasemia y Distribución del Agua del Cuerpo en un Niño Anurico

Se reporta un caso de una niña de 7 años de edad con una insuficiencia renal aguda la cual desarolló un aumento progresivo del potasio plasmatico asociado con alteraciones electrocardiograficas. Esta hiperpotasemia fué atribuida en parte a la anuria, y en parte a la gran concentración del potasio contenido en el jugo de naranjas administrado. La distribución del agua del cuerpo fué estudiada usando la técnica del "deuterium oxide" in vivo, el agua extra-celular mediante el metodo de la inulina y el volúmen del plasma fué determinado con el T 1824. Estos estudios revelaron que el volumen del plasma no se alteró y que el agua total del cuerpo fué esencialmente normal; sin embargo, se observó un aumento significante del compartimiento extra-celular con una disminución del compartimiento intra-celular.

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EFFECT OF TETRAETHYLAMMONIUM CHLORIDE (ETAMON CHLORIDE) ON MUSCLE SPASM IN POLIOMYELITIS

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THE constant search for improvement and simplification of the present accepted form of treatment of muscle spasm in the acute poliomyelitis patient prompted the authors to repeat the work of Lannon and Braudo[1] when tetraethylammonium chloride (etamon chloride*) was made available in June 1949. The tetraethylammonium ion has been shown by Acheson and Moe[2] to act as a blocking agent in the autonomic ganglia.

Though the total number of patients (259) admitted to this hospital in 1949 with the diagnosis of acute poliomyelitis far exceeded that of any previous year, many patients were found to be unsuited to this particular study, as they had either respiratory, bulbar or encephalitic symptoms. None of these patients was placed on tetraethylammonium chloride as an adverse course of events in these more complicated cases could not easily be attributed to the disease and absolutely dissociated from the therapy. Patients whose spasm was early, transient and unassociated with the ultimate development of true muscle weakness were not included in the study. No further selection of cases was made. At times not all patients available were used since detailed observations could not be recorded on more than five patients at a time.

Preliminary work was done with tetraethylammonium chloride, administering it intramuscularly and intravenously in varying amounts. The patients complained of moderate local pain when the medication was given intramuscularly. Adequate relaxation for physical therapy was obtained with 7 mg./kg. intravenously and, therefore, a greater dosage was not attempted. The intravenous route with a standard dosage of 7 mg./kg. was thus chosen for the study. The transient side effects previously described were noted in these patients whenever the medication was given. All experienced diplopia, loss of accommodation, tingling parasthesias, metallic taste in the mouth, and ptosis. An occasional patient experienced nausea. These symptoms usually were present within five minutes following injection and remained for 45 to 60 minutes. Since all were bed patients, no one complained of weakness or tiredness following injections. Tetraethylammonium chloride was given in the morning and again in the afternoon. A senior extern was in constant observation for several hours after administration of the medication, noting subjective comments and recording blood pressure determinations. Prior to the administration of tetraethylammonium chloride the mean blood pressure recording was 116/76 mm.Hg; within five minutes the mean recording was 108/74 mm.Hg. Thereafter there was a gradual return to normal over the next two hours.

The control group received warm, moist packs which were applied by the nursing staff

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in the currently accepted manner. Each patient received four packs daily except on Sunday. None of the patients was packed during the night.

Twice daily both groups received physical therapy consisting of muscle stretching to the point of tolerance. With few exceptions the same therapist was involved throughout the entire study, so that the degree of stretching was more nearly constant. Measurements were obtained by only one of the authors (F.E.) to avoid variation in determining the range of motion. Changes in the range of motion of the joints studied were measured in angular degrees with a goniometer. Patients were not included in the study until they had become afebrile, at which time the first goniometer measurement was taken. Subsequent measurements were made at 1 to 6 day intervals. Observations were terminated at times

TABLE 1

HIP FLEXION—PACK GROUP

Patient	Total angular degrees of improvement	Total days studied	Average daily improvement in angular degrees	d^2
1	15	15	1.00	0.0196
2	0	11	0.00	1.2996
3	25	15	1.67	0.2809
4	15	18	0.83	0.0961
5	10	18	0.56	0.3364
6	10	11	0.91	0.0529
7	10	11	0.91	0.0529
8	35	12	2.92	3.1684
9	10	11	0.91	0.0529
10	15	15	1.00	0.0196
11	20	12	1.67	0.2809
12	20	15	1.33	0.0361
$N_1=12$			$\bar{X}_1=1.14$	$\Sigma d_1^2=5.6963$

somewhat prematurely for lack of bed space and the necessity for transferring the patients to a convalescent center.

Convulsions occurred in one patient who had been given tetraethylammonium chloride. He received intramuscular medication for two days in the preliminary study, following which he received intravenous tetraethylammonium chloride twice daily, being given 7 mg./kg. During the evening of the fifth day he had two mild generalized convulsions lasting 2 to 3 minutes and 30 seconds, respectively. He was removed from the tetraethylammonium chloride series and, though his improvement was impressive, the information was not included in the statistical analysis because of the short duration of the observations. He received packs throughout the remainder of his hospitalization. An electroencephalogram recorded five days after the convulsions consisted of an irregular 7 to 9/second activity with many bursts of hypersynchronous 3 to 5/second waves. These findings suggest that the slow activity was due to a diffuse cerebral disorder. The patient complained of headache, vomited frequently, and was lethargic for nine days. Thereafter his convalescence was uneventful. An EEG seven months later was normal.

Seventeen patients were studied in detail. Comparative observations were made on 13 different functions with a varying number of patients occurring in each group, since all

did not have equivalent involvement. A total of 232 muscle functions were thus evaluated. Observations were recorded for hip flexion, knee flexion, hip adduction, hip abduction, dorsiflexion, knee extension, elbow extension in the neutral position, elbow extension with pronation, shoulder abduction, elbow extension with supination, shoulder flexion with the elbow flexed, shoulder flexion with the elbow extended, and wrist extension. It was not possible to devise an accurate means of recording variations in the spasm of the neck and back muscles.

An example of the manner in which the data were recorded is given in table 1. In this group where hip flexion was studied using hot packs there were 12 ($N_1 = 12$) patients. The average daily improvement in angular degrees was 1.14 ($\overline{X}_1 = 1.14$). The sum of the square of the deviation of the average daily improvement for each patient from

TABLE 2

t AND P VALUES FOR FUNCTIONS STUDIED

	t	$\overline{X}_1$	N_1	$\overline{X}_2$	N_2	P
Hip flexion	0.1209	1.14	12	1.19	17	> .90
Knee flexion	0.8236	1.49	11	1.05	17	> .40
Hip adduction	0.5889	1.11	10	1.40	19	> .50
Hip abduction	0.0239	0.82	12	0.83	16	> .90
Dorsiflexion	0.3803	0.34	13	0.26	15	> .70
Knee extension	0.0223	1.04	10	1.03	18	> .90
Elbow extension (neutral)	1.1515	2.12	2	0.89	3	> .30
Elbow extension with pronation	0.4938	1.67	2	1.05	3	> .60
Shoulder abduction	1.9224	0.92	8	2.52	4	> .05, < .10
Elbow extension with supination	1.6456	2.02	2	0.67	3	> .10
Shoulder flexion (elbow flexed)	0.7804	1.32	9	2.62	3	> .40
Shoulder flexion (elbow extended)	0.2915	1.65	6	2.05	3	> .70
Wrist extension	0.2443	0.82	9	0.98	5	> .80

the mean of the daily improvement for the group was 5.6963 ($\Sigma d_1^2 = 5.6963$).

The data obtained were subjected to the Fisher t test using the following formulae:

$$\sigma = \sqrt{\frac{\Sigma d_1^2 + \Sigma d_2^2}{N_1 + N_2 - 2}}$$

$$t = \frac{|\bar{x}_1 - \bar{x}_2|}{\sigma} \sqrt{\frac{N_1 N_2}{N_1 + N_2}}$$

Σd^2 = the sum of the square of the deviation of the average daily improvement for each patient from the mean of the daily improvement of the group.

N_1 = the number of patients in the pack series.

N_2 = the number of patients in the tetraethylammonium chloride series.

$\overline{X}_1$ = the average daily improvement in the pack series.

$\overline{X}_2$ = the average daily improvement in the tetraethylammonium chloride series.

The t values are given in table 2. The t values were subjected to the Fisher Probability Table for this value, and the P value was thus obtained. The t values obtained for all

functions fell below 2. That for elbow extension (neutral) was 1.1515, for shoulder abduction, 1.9224, and for elbow extension with supination, 1.6456. When compared with the P value the only one of possible significance was that of shoulder abduction with a value of $>.05$ and $<.10$. This gave preference to the etamon chloride group over the pack group. From the other figures one could not say that the etamon chloride group was either better or worse than the pack group, since the t values ranged from .0223 to .8236, and the P values ranged from $>.10$ to $>.90$.

Discussion

From a statistical analysis of the data one can not draw categorical conclusions from the study, as the total number of patients involved was not sufficiently great. However, one can say that there was no significant difference between the results of the two groups. If spasm in the neck and back could have been recorded, a more complete evaluation of the two methods of treatment would have been forthcoming. The impression was that this spasm did not respond well to either form of therapy, and that its resolution, therefore, becomes of significance in such a study.

Lannon and Braudo reported dramatic results with the use of tetraethylammonium chloride, particularly when it was given in amounts greater than the recommended 7 mg./kg. intravenously. In this series that single dosage was not exceeded. Additional data not reported in detail here were obtained to show that increasing the total daily dosage by more frequent administration of the medication did not produce significant variation in the reduction of spasm. Perhaps the discrepancy in the resolution of spasm observed by the present investigators and that observed by Lannon and Braudo can partially be explained by the fact that all patients included in this series were sufficiently involved to be ultimately paralytic.

Certainly the ward personnel preferred the tetraethylammonium chloride routine to the pack routine, for it was time-saving for the nurses. Many patients preferred the intravenous medication to packing, and when they were placed on pack routine preparatory to transfer to the convalescent home, they voluntarily requested return to the intravenous treatment.

Summary

A series of 232 muscle functions were carefully evaluated on 17 patients, comparing the results obtained in the reduction of spasm using the intravenous administration of tetraethylammonium chloride (etamon chloride) with frequent stretching as opposed to the conventional hot packs with frequent stretching.

The results were not statistically significant in showing the virtues of one form of therapy as opposed to the other. The subjective impression of the personnel involved and the preference of the patients for the medication make it seem reasonable to enlarge the series studied to determine its true place in the current poliomyelitis therapy.

Acknowledgment

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Addendum

In view of the recent British findings regarding pertussis and diphtheria vaccine injections and paralytic poliomyelitis, the cases reported here were again reviewed. It should

be remembered that this study is based on intravenous administration of tetraethylammonium chloride. The intramuscular route was abandoned in the preliminary work, as it was somewhat painful, and greater relaxation was obtained with the intravenous administration. In no instance was there noted a permanent increase in involvement. There was the casual patient who showed increased spasm on single observations, but the over-all picture was always one of improvement.

References

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Spanish Abstract

Efectos del Tetraethylammonium Chloride (Etamon Chloride) en el Espasmo Muscular de la Poliomielitis

Cloruro de tetraethylammonium, que actua como un agente inhibitorio de los centros (ganglia) autonomicos, se le administró a niños con poliomielitis paralítica al principio de la convalescencia.

La propiedad de este medicamento en aliviar el espasmo muscular y por consiguiente de modificar los cambios en la extensioń de los movimientos medidos en grados angulares mediante el uso de un goniometro; fué comparada con los resultados obtenidos en pacientes tratados con compresas calientes de uso convencional.

Los resultados obtenidos en este estudio no fueron suficientes en demostrar las virtudes de un metodo sobre el otro. La impresión subjectiva del personal implicado en este estudio y la preferencia de los pacientes por esta medicación sugiere aumentar la serie de casos estudiados, para así poder determinar su valor correspondiente en la presente terapia de la poliomielitis.

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HYPERTENSION IN POLIOMYELITIS WITH RESPIRATORY PARALYSIS

Report of Protracted Case With Necropsy Findings

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HYPERTENSION complicating poliomyelitis is known to occur and the present authors have observed it on six occasions in patients with respiratory involvement. With the exception of the recent survey by Grulee and Panos,[1] only a few isolated cases have been mentioned in the American and foreign literature. Salus[2] reported a case of bulbar poliomyelitis in which the systolic pressure was 200 mm.Hg five days after the onset of the disease. He also found transient hypertension in two cases of ascending paralysis. Inflammatory changes in the brain stem, particularly in the region of the formatio reticularis, were considered to be factors in the pathogenesis of the hypertension. In none of these cases was the hypertension sustained.

Reports on cases of poliomyelitis in which death occurred after a protracted course are rare. As Peers[3] has pointed out, most patients either die acutely or survive the initial attack. Consequently the neuropathologist sees either the acute changes, or occasionally the residua years after the disease process has become quiescent. Peers[3] reports three cases with respective survival periods of 7, 15 and 18 weeks.

This report concerns a case of sustained hypertension complicating acute poliomyelitis with a protracted course of 24 weeks. The neuropathologic findings indicate that the disease process was active up to the time of death. Thus, this case, as far as is known, is the longest reported period of survival of a patient still in an active phase of the disease.

CASE REPORT

The patient, a 16 yr. old white male, was admitted to Michael Reese Hospital with the diagnosis of acute anterior poliomyelitis. One week before admission, on a routine physical examination performed because his younger brother had contracted bulbar poliomyelitis,* blood pressure, urine and general physical condition were found to be normal; moreover, he had been actively engaged in strenuous varsity sports in high school, where repeated physical examinations as prerequisites for these activities had revealed nothing abnormal.

On admission temperature was 38.0°C. rectally and blood pressure 130/82 mm.Hg. His only complaints were headache, nausea and slight nuchal rigidity. Spinal fluid findings were: Pandy 4+, Ross-Jones 4+, cell count 700 with 60% neutrophiles and 40% lymphocytes. The following day his neck and back became stiffer, and positive Kernig and Brudzinski signs were noted. The uvula deviated to the right and the right side of the tongue was moderately elevated. These signs disappeared within 3 days.

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* This brother eventually recovered except for a residual partial paralysis of the palate.

TABLE 1

SUMMARY OF PERTINENT LABORATORY DATA

	Week		1	2	3	4	5	6	7	8	9	10	11	12	13
Urinalysis	Specific Gravity		1.008	1.024	1.022	1.021	—	—	—	—	—	—	1.010	1.009	1.008
	Albumin		±	±	±	+	±	0	±	±	+	0	±	+	+
	Hyaline Casts		0	Occ.	Occ.	3–4	2	Occ.	9	0	6	0	3	4	8
	Granular Casts		0	Occ.	0	0	3	0	2	0	0	0	0	2	Occ.
	Cylindroids		0	2	0	0–1	0	0	0	0	4	3	2	2	0
	WBC/hpf		2–3	1–5	0	1–2	7	0	9	3	11	13	15	25	45
	RBC/hpf		0	0	0	0–1	1	2	Occ.	0		1	Occ.	3–5	3
	Volume cc./24 hr.		—	—	—	—	—	1800	2600	2300	2850	1900	2900	3700	—
	Staph. Albus Culture		—	—	—	—	—	—	+	+	0	0	0	0	0
		Normal values													
Blood Chemistry	Urea N	(12–15 mg./100 cc.)	—	—	—	—	—	22	24	—	—	—	—	—	—
	Serum Ca	(9.5–11 mg./100 cc.)	—	—	—	—	—	—	13.1	—	—	—	—	—	—
	Serum PO_4	(3–4 mg./100 cc.)	—	—	—	—	—	—	4	—	—	—	—	—	—
	Blood Cl	(100 mEq./l.)	—	—	—	—	—	—	—	97.5	—	—	88.5	—	91.5
	CO_2 Combining Power	(21–31 mm./l.)	—	—	—	—	—	—	—	31.7	—	—	26	—	31
	Blood pH	(7.4)	—	—	—	—	—	—	7.3	—	—	—	7.39	—	—

The fever continued for 5 days, during which time the patient developed paralysis in the lower and then the upper extremities. The intercostal and abdominal muscles and the diaphragm were involved. He was treated symptomatically, placed in an Emerson respirator and given 300 cc. of poliomyelitis convalescent serum intravenously. In addition, 300,000 units of crystalline penicillin were given daily prophylactically.

Coincident with abdominal muscle involvement, there was loss of bladder and bowel sphincter control. This necessitated catheterization of the urinary bladder on the third through the sixth hospital day, when voluntary control was re-established. After subsidence of the fever, applications of hot packs were begun.

Other medication included oral prostigmine to promote bladder and bowel functions and supplementary vitamins. The patient received 2 gm. of salt daily to compensate for excessive perspiration. On the ninth hospital day, it was possible to remove the patient from the Emerson respirator and to place him in a Blanchard chest respirator, in which he was comfortable without cyanosis or tachycardia. Two weeks after admission, sufficient power had returned in the respiratory muscles to allow him to be taken out of the respirator for several hours at a time. Respiration at this time was almost entirely diaphragmatic with the aid of the nuchal accessory muscles of respiration.

On the 34th hospital day, his blood pressure was found to be 170/120 mm. Hg. Thereafter, the pressure varied from 190/120 to 170/100, finally stabilizing at about 180/120. The blood pressure, pulse rate and electrocardiogram (which was essentially normal) did not differ significantly, regardless of whether the patient was in or out of the Blanchard respirator. Except for his respiratory difficulty and generalized peripheral paralysis, he was relatively asymptomatic. Repeated urinalyses showed specific gravities up to 1.024 with traces of albumin and occasional hyaline and finely granular casts (table 1). The latter elements of the urinary sediment were at times as numerous as 10 to 12/hpf with increasing numbers of cylindroids, 1 to 5 white blood cells/hpf and an occasional RBC. Cultures of the urine revealed Staph. albus on several occasions.

Roentgenograms of the long bones showed moderate osteoporosis of the wrists and slight demineralization of the proximal end of the tibias at the epiphyseal line, apparently the result of skeletal atrophy of disuse. Intravenous pyelography on the 59th hospital day disclosed the urinary tract to be roentgenologically normal with normal excretory function.

Subsequent urinalyses disclosed the presence of pyelonephritis as shown by increasing numbers of hyaline, finely and coarsely granular casts, and many clumps of white blood cells in the urine. The specific gravity remained below 1.010, the urine volume varying between 1800 and 3700 cc. in 24 hr. Urine cultures became negative after penicillin and sulfathiazole were given; however, the character of the urinary sediment remained essentially unchanged. The patient's blood pressure persisted at about 180/120 mm.Hg.

Special Tests

A. Renal Studies.—On the 67th hospital day, endogenous creatinine and paramino-hippurate clearances were done revealing the following data (table 2): glomerular filtration rate 104 cc./min.; renal plasma flow 468 cc./min.; filtration fraction 22.2%. Simultaneous chloride clearances revealed normal chloride excretion.

B. Concentration-Dilution Test.—On the 102nd hospital day, maximum dilution was only 1.008, and maximum concentration after 15 hr. of withholding fluids was 1.010. The following morning the patient had several convulsions. The blood pH at this time was found to be 7.05, the serum chlorides 79.5 mEq./l., and the carbon dioxide combining power 32.8 m.Eq./l. The convulsions were attributed to an uncompensated acidosis, resulting from respiratory and metabolic dysfunction (manifested by the low blood pH, low serum chlorides, and the [apparently] normal carbon dioxide combining power). Thus, with the prolonged withholding of fluids to obtain maximum concentration of the urine, the kidneys were apparently unable to excrete accumulations of fixed acid metabolites. The carbon dioxide retention was thought to be due to the respiratory paralysis, for had there been no such paralysis, the acidosis (pH 7.05) probably would have caused hyperventilation with a resultant lowered carbon dioxide combining power. After forcing fluids and transferring the patient back to the large Emerson respirator, recovery from this episode was complete.

Four 24 hr. endogenous creatinine clearances done on the 135th through the 139th hospital days revealed a depressed glomerular filtration rate (mean glomerular filtration rate of 63 $\pm$ 7.6 cc./min.). Sodium and chloride clearances were not grossly abnormal at this time.

C. Hypoxia.—Because of the possibility that hypoxia due to respiratory paralysis might be the cause of the hypertension, blood oxygen and carbon dioxide were determined when the patient was out of the respirator. The arterial blood contained 18 vol. per cent of oxygen with 97% saturation and 55 vol. per cent of carbon dioxide. The A-V oxygen difference was 4.3 vol. per cent. Blood pH was found to be 7.3. These values are within the normal ranges.

D. Tetraethylammonium Chloride.—On the 80th hospital day the patient was given tetraethylammonium chloride 350 mg. to determine what role the sympathetic nervous system might be playing in producing the hypertension. After establishing a control level of blood pressure for 30 min., the drug was given intravenously. Within a few seconds the blood pressure had dropped from a level of 180/110 to 136/78 mm.Hg. The lowest systolic and diastolic readings were

TABLE 2

CLEARANCES

A. Routine Clearance 11/23/48 (67th Hospital Day)

Period	Duration (min.)	Urine flow* cc./min.	Creatinine clearance cc./min.	PAH† clearance cc./min.	Filtration fraction %
1	12	3.5	94.5	420	22.5
2	12	3.8	100	418	23.9
3	13	5.1	118	567	20.8
Average	—	—	104	468	22.2

B. Chloride Clearance‡ 11/23/48

	Plasma chloride mEq./cc.	Creatinine clearance cc./min.	Chloride filtered mEq./min.	Urine flow§ cc./min.	Urine chloride§ mEq./cc.	Chloride excreted mEq./min.	Chloride reabsorbed mEq./min.	% chloride reabsorbed	% chloride excreted	Chloride reabsorbed/l. H_2O reabsorbed
(1)	0.103	94.5	9.68	5.5	0.0513	0.28	9.40	97.1%	2.9%	103.0
(3)	0.104	118.0	12.30	8.1	0.0444	0.36	11.94	97.0%	3.0%	105.5

* Corrected for bladder wash.
Hydration:—oral water = 0.9% NaCl intravenously, 6 cc./min.
Average hematocrit—46%.
Average plasma protein—5.03 gm./100 cc.
† PAH = para-amino-hippurate.
‡ Chloride excretion studied during infusion of 0.9% NaCl at rate of 6 cc./min.
§ Uncorrected for bladder wash.

94 mm. Hg and 62 mm.Hg, respectively. Within 16 min. the diastolic pressure was back to the base line, reaching 118 mm.Hg at the end of 40 min.; the systolic pressure was re-established at 170 mm.Hg.

E. Sweating Test.—An iodine-starch preparation applied to the body was used to determine the presence or absence of autonomic nervous system activity in the skin, and indirectly the effect of polio virus on this system. The usual adjuvants of aspirin, hot fluids, multiple blankets, and heat cradle were employed to promote sweating. There was revealed moderate to marked blue color changes from the shoulders down to the ankles, equal bilaterally, indicating active responsiveness of the sympathetic nervous system. The head and neck were relatively unaffected.

COURSE

The patient's course continued unchanged until the 168th day after admission when suddenly, without warning, he started convulsing again, going into a deep coma. The blood pressure rose to 260/140-160. The therapy given previously was repeated but to no avail. The patient died 8 hr. after the onset of this episode.

Autopsy Report

The autopsy was performed 1½ hr. after death.

A. Brain and Spinal Cord

Gross Examination: The brain was edematous and weighed 1450 gm. The spinal cord markings were indistinct and blurred at all levels and in both anterior and posterior horns. The tissues were fixed in formalin. Sections for microscopic examination were taken from various regions of the cord, spinal sympathetic ganglia, medulla oblongata, mesencephalon, basal ganglia and cortex. Celloidin sections were stained with toluidine blue, Lillies' azure-eosin, hematoxylin-eosin, and with myelin stains.

Microscopic Examination: Type of changes seen: (1) Inflammation, as exhibited by cuffing of blood vessels by small round cells; (2) loss of nerve cells as well as fibres; (3) proliferation of microglia, diffuse in some areas and focal in others; (4) marked proliferation of astroglia, the most prominent lesion in all levels of the spinal cord; (5) small focal hemorrhages without reaction of surrounding tissue.

Distribution of Lesions: 1. Spinal cord.—The lesions found in the spinal cord were more or less uniform and consisted of almost complete disappearance of motor cells of the anterior horn. The lateral horns were severely involved. The posterior horns exhibited slight loss of nerve cells. Clark's columns appeared well preserved. Acute inflammatory changes were insignificant although occasional vessels were surrounded by round cells. Microglial proliferation was almost absent. Myelin sheath stains revealed almost complete loss of fibres and the same to a minor degree was true for the posterior tracts, especially for the region of Schultze's comma. In addition, the cervical cord showed 2 foci measuring approximately 2 x 1 mm., consisting of loose edematous tissue containing small astrocytes and scanty fibres surrounded by large "fattened" astrocytes.

2. Medulla.—The lesions of the medulla differed from those in the cord in that inflammation and proliferation of microglia were prominent and proliferation of astroglia practically absent. The lesions were widespread and diffuse. The most severely affected regions were the reticular formation and the nucleus ambiguus; round cell infiltration and proliferation of microglia were marked, and necrotic nerve cells were more frequent here than in any other region of the central nervous system.

3. Less severe, but also marked changes were found in the nucleus of the 12th nerve, and in the dorsal motor nucleus of the vagus. Less affected were the spinal nucleus of V, nucleus gracilis and cuneatus and the olives.

4. Fairly marked changes were found in the red nuclei.

5. Deiters' nucleus, the substantia nigra and the central gray matter around the iter exhibited minimal changes.

6. Cerebellum, cerebellar nuclei and pontine ganglia, basal ganglia, and internal capsule were spared.

7. The hypothalamic region revealed dilation of blood vessels and distention of perivascular spaces to a marked degree.

8. Numerous sections from the cortex revealed occasional small hemorrhages and distended perivascular spaces; otherwise the architecture was well preserved.

9. Degeneration of the anterior roots was marked in all sections of the cord.

10. Examination of the spinal sympathetic ganglia showed occasional pyknotic dark nerve cells but no inflammation.

B. Kidneys and Other Organs

Gross Examination: 1. The kidneys weighed 340 gm. and were moderately swollen. The capsules stripped with ease, revealing pale yellow and tan surfaces with accentuation of fetal lobulation. Cut section revealed a deep purplish red medulla in distinct contrast with a pale tan cortex. Along the periphery of the cortex there were some areas of flattened scars.

Microscopic Examination: The glomeruli were large and irregular. A basophilic substance was noted between the glomerular tufts and the basement membrane. Occasional glomeruli appeared atrophied. Moderate congestion of the glomerular capillaries was present. The tubules were distorted and irregular. In some areas there was evidence of tubular atrophy, in others degeneration. In the lumina of many tubules were dark basophilic, calcific substances. In these tubules a large

number of lymphocytes and a few polymorphonuclear leucocytes were seen. In addition, there were several multinucleated giant cells. The proximal convoluted tubules were more severely involved than the others. The interstitial tissue contained large collections of lymphocytes and occasionally a few polymorphonuclear leucocytes. There was an increased amount of fibrous connective tissue in these areas. The smaller arteries showed an intimal proliferation and thickening, so that the lumina of these vessels were severely narrow. Arterioles showed no changes. Diagnosis: marked subacute pyelonephritis with calcific deposit in tubules.

2. The heart weighed 280 gm., showed moderate hypertrophy of the left ventricle and was otherwise grossly normal. Microscopic sections revealed a moderate subacute myocarditis with some patchy fibrosis and fatty infiltration.

3. The right lung weighed 320 gm. and the left 270 gm. Patchy areas of bronchopneumonia and emphysema were seen. The tracheo-bronchial tree was not remarkable.

4. Cloudy swelling and passive hyperemia of the spleen, kidneys and liver were present.

Discussion

The development of sustained hypertension during the protracted course of acute poliomyelitis in this case focuses attention on the possible pathogenetic roles of the kidneys and the central nervous system.

In consideration of the central nervous system two features warrant particular discussion: (1) the protracted course of the disease and (2) marked damage in the lateral horns of the spinal cord and in the so-called vasomotor centers in the medulla.

The pathologic findings in the brain stem and spinal cord clearly reflect the long duration of the disease. The spinal cord showed the astroglia scars, with a few traces of an active process, whereas the medulla oblongata and the mesencephalon exhibited findings identical with those which are common in acute poliomyelitis. It seems justified to conclude that in this patient the disease remained active for a considerable period. In other words, poliomyelitis is not always an acute disorder which either kills quickly or is survived with residua; rather it may assume a subacute or chronic course and in occasional cases may never become inactive.[4]

This case seems to offer at least suggestive supportive evidence for the assumption that the morphologically demonstrable lesions in the brain stem and lateral horns of the spinal cord played a contributing role in either the etiology or maintenance of hypertension. The regions in the brain stem which were affected most severely were the nucleus ambiguus and the reticular formation. The Minnesota Poliomyelitis Research Group[5] found intense changes in the dorsal reticular substance in cases with severe vasomotor disturbances, and the pathologic studies of Baker et al.[6] further corroborates this observation. Experimental work[5,7] also indicates the presence of a vasomotor center in the reticular formation. Injury and experimental stimulation of this region have produced sweating and hypertension.[8–11] The recent observations by Chor and Zivin[12] on the vasopressor sensitivity to peripheral and bladder stimuli in patients with high spinal lesions bear close resemblances to the similar observation that hypertension is not uncommon in poliomyelitis with high spinal or medullary involvement. In the patient reported here, the sweating activity starting from the shoulder and reaching the ankles, as demonstrated by the iodine-starch test, indicated the presence of intact sympathetic activity. This is unlike the usual course in poliomyelitis, where there is progressive decrease in sweating. As with the iodine-starch test, the marked drop in blood pressure with the administration of tetraethylammonium chloride further enhances the evidence for excessive sympathetic tone.

The lesions of the kidney apparently developed and progressed *pari passu* with the

progressive disease in the central nervous system. It is not uncommon for severe cases of poliomyelitis to develop renal complications such as calculi and infections. Since repeated physical examinations and urinalyses prior to admission were normal, it is reasonable to assume that neither hypertension nor advanced renal disease existed prior to the present illness. Stasis of urine, coupled with catheterization, probably initiated the infection in the kidney. When the hypertension was first discovered, both granular and hyalin casts with a constant trace of albumen had been present for several days. Urine concentration and dilution test were within the normal range. However, paraminohippurate clearance showed decreased effective renal blood flow. With a normal filtration rate it was assumed that there was efferent arteriolar vasoconstriction resulting in an increased filtration fraction. At this stage in the disease the increased sympathetic outflow with diffuse vasospasm, probably involving the renal efferent arterioles, may have been a sustaining mechanism in the elevated blood pressure. However, Fishberg[13] and Weiss and Parker[14] maintain that during the acute phase of pyelonephritis, such as the present authors assume this patient was in at this time, hypertension is exceedingly uncommon. Subsequently, pyelonephritis was definitely established clinically, and renal function became grossly impaired. The dilemma as to whether the hypertension was sustained at this time by the irritable vasomotor apparatus or the disease of the kidney remains unresolved.*

It seems probable, however, that since the hypertension had reached a fairly stable level before marked urinary changes were noted, that the alterations in the vasomotor center and lateral horns played at least a contributory role in the etiology and sustenance of the hypertension in this case.

Summary and Conclusions

A case of sustained hypertension complicating acute protracted poliomyelitis with respiratory involvement is presented. Data of studies of the autonomic nervous system and the kidneys are given. Complete necropsy findings, including detailed examination of the central nervous system, are presented. The role played by the nervous system and kidney is discussed, and it is suggested that in this case the hypertension probably was due to neurogenic factors.

Acknowledgment

The authors are indebted to other members of the Cardiovascular Department, especially Drs. L. Akman and J. Stamler, for their assistance in collecting the data. They are also grateful to Drs. L. N. Katz, H. Salans, O. Saphir and R. Sternheimer for their helpful suggestions.

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* The role of hypoxia in the production of experimental hypertension is highly controversial. Evidence in man is lacking. Arterial O_2 and CO_2 determinations and arteriovenous O_2 difference were within normal limits in the case. and while it is conceivable that the above determinations may be within normal limits with the existence of local hypoxia because of edema, markedly increased local demands, etc., the hypothesis that anoxia was responsible for the hypertension in this case is untenable with the evidence at hand.

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SPANISH ABSTRACT

Hipertensión en Poliomielitis con Paralísis Respiratoria; Reporte de un Caso con Estudio Necropsico

Los autores describen un caso que presentó una hipertensión arterial persistente durante el curso de una poliomielitis aguda prolongada con paralísis respiratoria. Se presenta data de las observaciones obtenidas mediante el exámen del sistema nervioso autonomico y renal. Se describen los hallazgos del exámen necropsico, incluyendo un estudio detallado del sistema nervioso central. Los autores discuten el role desempeñado por el sistema nervioso central y riñones, sugiriendo que la hipertensión en este caso fué probablemente debida a factores neurogenicos.

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PNEUMATOSIS INTESTINALIS

Review of the Literature with Report of 13 Cases

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PNEUMATOSIS of the intestine (also called pneumatosis cystoides intestinorum hominis, intestinal emphysema, cystic lympho-pneumatosis, peritoneal pneumatosis, entero-mesenteric bullous emphysema, and gas cysts of the abdomen) consists of the presence of gas-containing cysts of varying size within the wall of the intestinal tract, sometimes accompanied by similar cysts on the surface of other abdominal viscera and parietal peritoneum. It is much discussed in European literature, but largely unknown in North America. Twelve infants and three older children were included in the world literature on the subject prior to a recent report of 17 cases in infants.[1]

Intestinal pneumatosis is better known in veterinary than in human medicine; it is common in pigs.[2-4] Duvernoy observed it during dissection of a cadaver in 1747.[5,6] Cloquet[7] in 1820 and Bang[8] in 1876 reported finding it at autopsy. Nigrisoli[9] and Hahn[10] independently reported finding pneumatosis at laparotomy; Finney[11] was the first to report it in this country. Reverdin,[12] in 1924, reported its pre-operative diagnosis, based on clinical and roentgenologic observations and confirmed at laparotomy. Lerner and Gazin[13] were the first Americans to report a nonoperative diagnosis, based on roentgenologic evidence. Though some of the older case reports are not accepted as true pneumatosis, over 200 acceptable cases have now been reported, from practically all civilized countries. Those concerning infants and children are summarized in table 1. Pneumatosis was found at operation in the first two patients listed; in all others it was an autopsy finding.

The reader is referred to the bibliography for more detailed discussion of pneumatosis intestinalis in adults. Much of the literature reviewed was found sketchy and inconclusive; only the reports which seemed of some value and reliability are included here.

The peak incidence of pneumatosis is between 25 and 50 years, but Lyons[22] observed it in a man aged 81, and case 13 of this series was a premature baby aged 12 days.

Two rarer conditions, pneumatosis vaginae[23,24] (colpohyperplasia cystica, vaginitis emphysematosa) and pneumatosis vesicae,[23] deserve mention because of their similarity to pneumatosis intestinalis. Vaginal and intestinal pneumatosis may coexist;[25] they are believed[23,26] to have the same pathogenesis.

Pathology: The lesion looks much like hydatidiform mole. The thin-walled cysts vary greatly in size;[23,27] in infants they are less than 1 cm. across, and sessile. When compressed, they move a short distance, or burst without collapse of surrounding cysts. Pinpoints or networks of serosal scars have been described near co-existent lesions or at sites of earlier lesions.[28-30]

The ileum is first, the colon second, in frequency of involvement; both may be affected at once. Other visceral surfaces are less commonly involved, usually along with

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TABLE 1

Author, Year	Sex	Age at death	Clinical picture, duration	Site of pneumatosis	Other GI lesions	Extra-intestinal disease
Viscontini[14] 1904	F	13 yr.*	Pyloric obstruction by mesenteric adenomas, recurrent after gastroenterostomy	d, j, n	Adhesions between stomach and mesocolon	Mesenteric adenomas, nature?
Urban[15] 1910	F	13 yr.†	Peritonitis at 5½ yr.; vomiting, abd. pains, distention—3 yr.; pneumoperitoneum, duration?	b, c, e, f	None observed at laparotomy	Pulmonary TBC
Müller[16] 1912	M	11 wk.	Vomiting since birth; hematemesis 2 days; melena, duration ?; anemia, duration?	a (rest of gut not examined)	Hypertrophic pyloric stenosis; injection and petechiae in pyloric area	? (autopsy restricted)
Schnyder[17] 1917	M	5 yr.	Vagal paralysis, diphtheritis myocarditis, duration?	d, j, k	None	Diphtheria
Moore[18] 1929						
case 1	F	9 wk.	Diarrhea 4 days; convulsions 4 hr.; antemortem hematemesis	b, c; k (microscopic)	Mucosal and submucosal hemorrhage; blood in gut lumen	Pneumonia RUL; thymic hyperplasia
case 2	F	14 mo.	Vomiting and diarrhea, 6 days	d, i (most marked in ascending)	Acute catarrhal enteritis; petechiae in jejunum and ileum	Pulm. congestion, emphysema; purulent mastoiditis; congestion spleen
case 3	M	8 wk.	Failure to gain; vomiting 7 wk.; acute otitis, duration?	b, c	Acute catarrhal enteritis; 5 mm. ulcer 15 cm. above ileocecal valve	Purulent otitis and mastoiditis; pulm. congestion, atelectasis, emphysema; fatty liver; edema brain
case 4	M	3 wk.	Diarrhea 2 wk.; skin rash, duration?	i	None noted	Pemphigus; patent foramen ovale
Botsford and Krakower[19] 1938						
case 1	M	6 wk. (premature)	Failure to gain; pneumonia, cyanosis >2 wk.	i	None noted	Suppurative bronchopneumonia; bronchial lymphadenitis
case 2	M	9 mo.	Nephrosis 3 mo.; URI, vomiting, diarrhea, 1 wk.	b, c, f, h, k, l, m	Distention of jejunum	Subacute glomerulonephritis; bronchopneumonia; emphysema; acute otitis; mild biliary cirrhosis; mesenteric lymphadenitis
case 3	M	8 mo.	Pneumonia 6 wk.; vomiting and diarrhea 2 wk.	c, k	Small recent ulcers in esophagus; pinpoint opening at proximal end of cysts	Bilat. bronchopneumonia; bilat. otitis; mesenteric adenitis; edema brain; fatty liver
case 4	M	3½ mo.	Diarrhea, vomiting, jaundice, enlarged liver, 2 wk.	b, k	Gastric distention	Pulm. congestion; obstructive biliary cirrhosis; mesenteric adenitis; mild rickets and scurvy

Author, Year	Sex	Age at death	Clinical picture, duration	Site of pneumatosis	Other GI lesions	Extra-intestinal disease
case 5	M	4 mo.	Vomiting, diarrhea, 17 days	c (antemesenteric edge)	Gastric distention; small intestinal mucosa congested, atrophic over cysts	Sl. bronchopneumonia; biliary cirrhosis; bilat. otitis; mesenteric adenitis‡
case 6	M	9 wk.	Epidermolysis bullosa, vomiting, diarrhea, melena, duration?	i	Aberrant pancreas in stomach; thrombi in mucosa and submucosa; mod. distention	Epidermolysis bullosa; bronchopneumonia; otitis, mastoiditis; mesenteric adenitis
Lindsay, Rice and Selinger[20] 1940	F (Negro)	7½ mo.	Diarrhea 2 wk.; rhinitis; positive Kahn	c, f	Many small ulcers in stomach; few thrombi in small vessels	Splenic fibrosis
Judge, Cassidy and Rice[1] 1949						
17 infants	M 10 F 7 (white 11, Negro 6)	1–8 mo.	Fever in all pts.; vomiting and diarrhea "prominent"; respiratory infection in 15 pts.; severe diarrhea in 2 pts. No RGs	b 5 c 12 e 10 f 7 g 2 h 1 j 1	Ulcerative ileocolitis in 13 pts.; various stages ileocolitis in 4 pts.; Salmonella infection in 2 pts.; hemorrhage, thrombosis and edema with variable inflammation	Not mentioned

Site:

a. stomach
b. jejunum
c. ileum
d. small intestine, segment not specified
e. cecum
f. ascending colon
g. transverse colon
h. descending colon
i. colon, segment not specified
j. mesentery
k. mesenteric lymph nodes
l. cisterna chyli
m. thoracic duct
n. parietal peritoneum

* Patient discharged in good condition.

† Short-circuiting anastomosis done at 2nd laparotomy; patient made slow recovery after repeated paracenteses of gas and fluid.

‡ Twin died with same complaints; autopsy showed no pneumatosis.

Maass[21] in 1904 reported a 7 mo. old male clinically diagnosed "pseudo-chylous ascites" who died of "heart failure" and at autopsy had a cystic mass in the omentum connected by a fistula to the stomach; this is not generally accepted as true pneumatosis and is not included in the table.

intestinal involvement.[23] Pneumatosis is often confined to an intestinal loop constricted by adhesions[12] or volvulus,[31-33] sometimes to a loop containing intestinal parasites.[32, 34, 35]

In infants, cysts are predominantly submucosal. The mucosa may show atrophy[36] or edema.[31] The cellular infiltrate varies, but usually indicates chronic inflammation; giant cells are rarely seen in infants. Gas has been observed in vessel walls[36] and nerve sheaths.[31] The cysts have been thoroughly described by Masson;[26] he believes them to begin in lymphatics, ending as a scar ringed by elastic fibers.

The gas is a mixture of 70-90% nitrogen, 3-20% oxygen, and 0-15% carbon dioxide.[15, 31] Urban alone found methane by paracentesis on a living patient.[15] These proportions closely resemble those found by Banyai 48 hours after induction of pneumoperitoneum with pure oxygen.[37]

Associated gastrointestinal disease is prominent, in adults mostly peptic ulcer with pyloric stenosis;[36] various other intestinal lesions, and cardiac and pulmonary disease, are mentioned.[23, 33, 38, 39] In infants, diarrheal disease most frequently coexists, pulmonary disease ranking next.

Pathogenesis: Little experimental work has been done relative to those disease. The theories discussed here are individually inadequate; probably all three factors operate in the production of cysts.

1. Infectious: Many organisms, including yeasts, have been isolated from cysts in living patients;[40] Jaeger's "Bacterium coli lymphaticum aerogenes" reproduced the disease in animals but killed them within 48 hours.[41] Killian[23] considers pneumatosis the end result of lymphangitis due to low-grade, nonspecific infection. Lymphangitis elsewhere produces no gas cysts, and it does not explain the lack of inflammation around cysts or the occurrence of extraintestinal pneumatosis.

2. Biochemical: Pneumatosis in pigs seems related to diet;[12, 26] such relation is not clear in man. Absorption of free or dissolved gas is supported by Stübel's observation of bubbles of oxygen formed in lymphatics of intestine placed in peroxide solution.[42] Masson blames acid mucosal secretions, plus circulatory stasis.[26] This does not explain extraintestinal pneumatosis, its association with peptic ulcer, or its occurrence where absorption is least active.

3. Mechanical: Air injected directly into the gut wall follows lymphatics to the mesentery and disappears within a few days.[31] Box saw emphysema around a peptic ulcer after washing it with air and water.[43] Miyake forced air into a gut loop after scraping the mucosa and waiting for exudate to cover it; the resulting blebs remained 5 to 6 days.[44] Tung and Ngai made an artificial stenosing peptic ulcer in a dog; when the stomach was filled with air and salt solution, air bubbles appeared inside and outside lymphatics, progressing distally with peristalsis, some remaining two days in serosa and omentum.[34] Menna[35] injected air into the gut wall of rabbits and found the gut normal 10 days later, but if gas withdrawn from the animal's intestine was injected, 10 days later there was increased density of the lymph follicles with microscopic dilatation of the surrounding subcapsular space. When a culture of B. coli was injected 20 days before air injection, small vesicles were visible in the peritoneum 15 days after air injection, plus adhesions and mesenteric thickening. Streptococci and staphylococci produced more adhesions. Giant cells were absent, though air injected subcutaneously is said to produce them within a few days.[19]

The most significant clinical observation was that of Tung and Ngai.[34] Their patient

suffered from stenosing peptic ulcer, pneumoperitoneum and pneumatosis of the small intestine. During laparotomy he vomited, and the cysts at the mesenteric border immediately swelled, without simultaneous distention of the intestine.

The mechanical theory does not explain why bacteria do not enter the intestinal wall, nor why pneumatosis is rare in obstruction, despite hyperperistalsis, distention and thinning of the mucosa.

Clinical Picture: Symptoms of primary pneumatosis refer only to adults;[29, 31, 38] they range from acute to chronic, often mimicking appendicitis, peritonitis or intestinal obstruction. The only significant sign is a crepitant, nontender abdominal mass. Pneumoperitoneum in a patient with peptic ulcer is said[20, 31] to be pathognomonic of pneumatosis if perforation can be excluded. Roentgenograms show pneumoperitoneum and cysts; interposition, either fixed or reversible, of small intestine between liver and diaphragm (the Moreau-Chilaiditi syndrome) is very suggestive.[29, 31, 45–50] Barium enema shows colonic cysts as translucent filling defects.[13,31,51]

Pneumatosis is generally of short duration, from 2 to 40 days; re-operations have shown only roughening and scarring of the serosa. However, it may persist a long time or recur.[34] Submucosal cysts may occlude the lumen or initiate an intussusception;[52] subserosal cysts may initiate a volvulus.[29] Interposition alone does not cause obstruction.

Treatment: None is indicated in the absence of intestinal obstruction. Simple laparotomy is said to have curative effects.[26] Closure without drainage suffices for primary pneumatosis; puncture or removal of cysts is unnecessary. Treatment of associated intestinal disease follows the usual indications; healing is normal after resection or anastomosis.

Material

Cases 1 and 2 of this series were observed by the writer in 1948 at Notre Dame Hospital, Montreal. Cases 3 to 12 were found in the records of more than 1800 autopsies performed in 18 years at the Children's Memorial Hospital, Montreal. Case 13 was observed by Dr. James B. Arey at Charity Hospital of Louisiana at New Orleans.

Case 1 (M.G., 1948), white female born of a pre-eclamptic mother after 7½ mo. gestation, was transferred to pediatric service at age 1 mo. a/c diarrhea and persistent rectal prolapse, with blood and pus in the stools. Stool cultures were negative. Pneumonia and otitis yielded to antibiotics. She died on the 50th day of life, showing fever, distention and dehydration.

Autopsy (4 hr. post mortem) showed: mastoiditis; interauricular septal defect; pneumatosis of the colon from ileo-cecal valve to sigmoid, excluding the appendix (Fig. 1).

Microscopic examination of the colon showed submucosal cysts lined with endothelium which in larger cysts resembled fibroblasts (Fig. 2). Lymphatics underlying a small mucosal ulcer were full of PMN. The nerve plexi were edematous with occasional necrotic ganglion cells.

Case 2 (R.B., 1948), white male aged 3 wk., was admitted a/c pneumonia of 4 days' duration. Diarrhea developed on the 5th hospital day, followed by fever, distention and intermittent vomiting. RGs taken on the 2nd and 10th hospital days showed on postmortem review a narrow radiolucent band outlining the distal colon (Fig. 3). He died on the 13th hospital day.

Autopsy (9 hr. post mortem) showed: closed retropleural cyst lined by gastric mucosa; bronchopneumonia and atelectasis; intramesenteric diverticulum beginning close to the pylorus and communicating with the jejunum 30 cm. distally; pneumatosis of the colon, exactly resembling Case 1.

Microscopic examination showed submucosal cysts, and mucosal abrasion, with edema, congestion, and infiltrate of PMN, lymphocytes and plasma cells (Fig. 4).

Case 3 (J.A., 1934), white male aged 3½ mo., was admitted a/c vomiting and athrepsia since the first few days of life. He died on the 73rd hospital day with dehydration, distention and otitis media.

Autopsy (36 hr. post mortem) showed: bronchopneumonia, low grade ileitis, and pneumatosis of the ileum and colon (examination of head not permitted).

Microscopic examination showed submucosal and a few subserosal cysts. The smaller cysts appeared to be lymphatic vessels without endothelial lining. The submucosa showed edema, vascular stasis, cellular infiltration, and irregular fluid-containing spaces, perhaps early cysts.

Case 4 (M.W., 1935), white female aged 7 wk., was admitted a/c diarrhea, vomiting, athrepsia

FIG. 1. Case 1.—Gross appearance of colon after fixing. Cysts were 1 to 5 mm. in diameter.

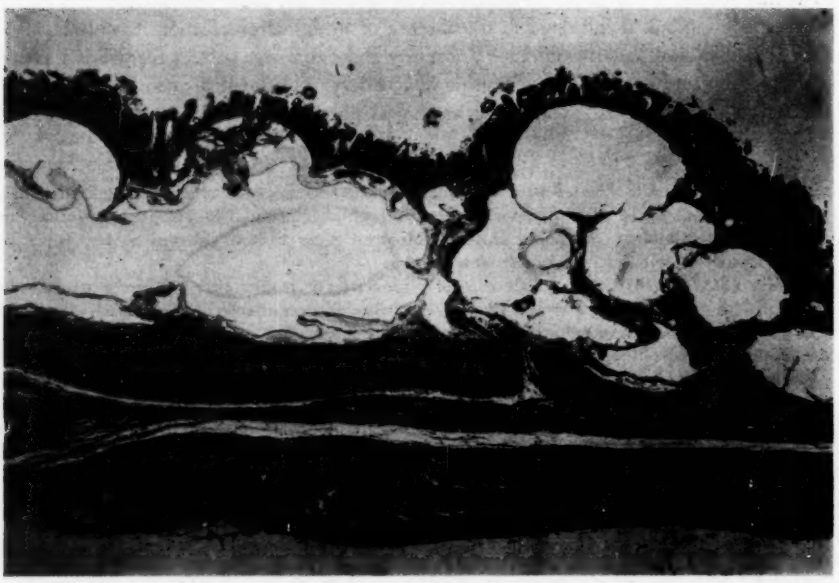

FIG. 2. Case 1.—Microscopic appearance of colon, showing characteristic appearance and submucosal situation of cysts.

and chronic otitis. She died on the 7th hospital day of aspiration pneumonia.

Autopsy (12 hr. post mortem) showed: marasmus; hemorrhagic alveolar pneumonia, probably streptococcal; pneumatosis of the cecum and 12 cm. of ascending colon. Mesenteric nodes were atrophic.

Microscopic examination showed numerous submucosal and occasional subserosal cysts, frequently endothelium-lined, with a few inflammatory and giant cells. The serosa showed edema and a thin

FIG. 3. Case 2.—Roentgenogram taken 11 days before death. Arrows indicate increased radiolucency outlining involved distal colon.

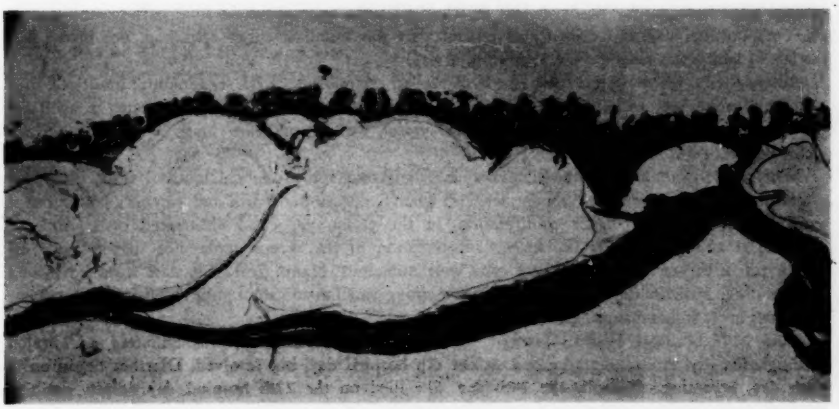

FIG. 4. Case 2.—Microscopic appearance of colon. Rings of pink-staining material within cysts are barely visible.

cellular infiltrate; many cocci present were considered postmortem invaders because of the lack of surrounding tissue reaction; the epithelium was intact.

Case 5 (V.L.M., 1936), white male aged 7 wk., was admitted a/c coryza for 2 wk. and pneumonia for 1 day. Distention was extreme by the 5th day; he died on the 11th hospital day.

Autopsy (5 hr. post mortem) showed: hemorrhagic alveolar pneumonia with middle lobe atelectasis; multiple lung abscesses and encysted empyema yielding Staph. pyogenes; cystic fibrosis of the pancreas; moderate atrophy of mesenteric nodes; and marked distention of small intestine and proximal colon, with 4 separate areas of pneumatosis, up to 5 cm. long, along the ileum.

Microscopic examination showed submucosal and mucosal cysts, surrounded by minimal focal inflammation; the intervening intestine was normal.

Case 6 (C.B., 1937), white male aged 4 mo., was admitted a/c vomiting and athrepsia. He became more toxic and dehydrated, and died on the 16th hospital day.

Autopsy (5 hr. post mortem) showed: underdevelopment; otitis media; mastoiditis; interstitial pneumonia; moderate atrophy of mesenteric nodes; and pneumatosis along 23 cm. of the ascending colon.

Microscopic examination showed minimal inflammation around the cysts except for a loculated abscess which resembled the cysts in outline and communicated with the intestinal lumen by 2 narrow sinus tracts lined with fibrino-purulent exudate. Some cysts were partially lined by endothelial-type cells; a few giant cells were seen in the cyst walls.

Case 7 (J.G., 1946), white male aged 2 mo., was admitted a/c progressive jaundice for 9 wk., weight loss, umbilical discharge and pale stools. Abdominal RG showed no sign of pneumatosis on later review. No pneumatosis was seen at cholecystectomy on the 9th hospital day. He grew worse from the 4th postoperative day, eviscerated on the 18th hospital day, and died following secondary closure.

Autopsy (22 hr. post mortem) showed: emaciation; marked icterus; plastic peritonitis between transverse colon and liver; congenital atresia of the bile ducts; a mucosal ulcer 4 cm. in diameter at the jejuno-ileal junction, containing 2 tiny fresh perforations, with pneumatosis extending distally from the ulcer to involve the entire ileum and colon except the appendix.

Microscopic examination showed the ulcer to extend to the muscularis mucosae, with many submucosal and mucosal cysts around the perforations. Large submucosal cysts above and below the ulcer contained PMN and giant cells; a deep ulcer, lined by fibrin, extended to the serosa and might have been a ruptured infected cyst.

Case 8 (Baby C., 1946), white male aged 1 day, was admitted a/c general weakness. Atelectasis and pneumonia yielded to routine therapy. He developed multiple staphylococcal abscesses at venepuncture and cut-down sites. He recovered well from Fredet-Ramstedt pyloromyotomy done on the 17th hospital day, and correction of malrotation of the colon done on the 38th hospital day. On the 63rd hospital day he developed diarrhea, with Ps. pyocyaneus on stool culture, and died on the 66th hospital day. No evidence of pneumatosis was found at either operation, nor in the RGs, the last of which was taken on the 38th hospital day.

Autopsy (20 hr. post mortem) showed: underdevelopment; RUL pneumonia; multiple intra-abdominal adhesions without peritonitis; and pneumatosis of the colon. In the cecum a small area of congestion surrounded a thrombosed mural vessel. Culture of heart, lungs and abdominal wall yielded Esch. coli.

Microscopic examination showed numerous submucosal and occasional mucosal cysts, many with a flat lining. Some contained monocytes and giant cells in the walls and lumina; collections of similar cells, with fibroblastic proliferation, in the cecum suggested obliterated cysts. Abundant submucosal and serosal foci of chronic inflammation in the cecum diminished distally to a few monocytes in the rectosigmoid, where cysts were numerous. Many giant cells were scattered through the intestinal wall, in the serosa perhaps representing reaction to catgut suture. The jejunum showed only early acute inflammation, giant cells and edema.

Case 9 (L.N., 1947), white female aged 1½ mo. (premature), was admitted a/c lifelong feeding difficulty. Atelectasis appeared on the 6th hospital day, but resolved. Diarrhea began on the 11th day, becoming severe by the 24th day. She died on the 27th hospital day despite antibiotic therapy.

Autopsy (17 hr. post mortem) showed: bilateral focal pneumonia; bilateral otitis media; minimal adrenal hemorrhage; fatty liver degeneration; old dural blood stains; and pneumatosis of the lower

ileum to the ileocecal valve, with retroperitoneal and mesenteric edema and scant hydroperitoneum. Microscopic examination showed submucosal cysts with minimal inflammatory reaction, and moderate atrophy of the mesenteric LN.

Case 10 (L.B., 1948), white female aged 11 mo., was admitted a/c persistent diarrhea for 6 mo. She grew steadily worse despite extensive Rx. On the 25th hospital day, blood appeared in stools and vomitus; stool culture yielded P. morgani. She died on the 34th hospital day.

Autopsy (8 hr. post mortem) showed: extreme emaciation; purulent otitis media; and acute ulcerative pseudomembranous ileitis and colitis with localized pneumatosis around the ileocecal valve, underlying an ulcer which was superficial, minimally indurated, and covered with mucoid substance; all other ulcers were indurated, raised and covered with adherent membrane. At the splenic flexure were a large ulcer, several hemorrhagic saccular protrusions, and small indurated black nodules.

Microscopic examination showed no acute or chronic reaction around the cysts. The mesenteric nodes showed only moderate atrophy. Extensive bacteriologic study revealed no dysentery organisms; the enteritis was believed to be staphylococcal.

Case 11 (N.C., 1948), white male born 5 wk. prematurely, had a pyloromyotomy at age 3 wk. and was discharged 9 days later in good condition. Vomiting and diarrhea began the next day; he was re-admitted at age 5 wk. He grew steadily worse; fresh and occult blood appeared in the stools on the 3rd hospital day; he died on the 5th hospital day.

Autopsy (14 hr. post mortem) showed: hypertrophy of pyloric muscle with adequate lumen diameter; ulcerative jejunitis, ileitis and cecitis; and pneumatosis, marked in lower jejunum and ileum, slight in the cecum where there were few ulcers.

Microscopic examination showed no cellular reaction around the cysts; there was moderate atrophy of the mesenteric LN. Elaborate postmortem cultures revealed no dysentery organisms; the enteritis was believed staphylococcal.

Case 12 (R.R., 1949), white male aged 6 yr., had been studied at another hospital a/c rapid weight gain. He arrived at the admitting office in coma, which followed indigestion and vomiting 4 days before, and heavy breathing the day before. He died a few minutes after arrival; the abdomen was markedly distended and tympanitic at death.

Autopsy (22 hr. post mortem) showed: marked obesity; otitis media and mastoiditis; skin petechiae; septicemia with meningococci and Str. pyogenes in the heart blood; and 2 patches of minimal pneumatosis of the jejunum without visible mucosal defect.

Microscopic examination showed no evidence of inflammation, and only moderate atrophy of the LN.

Case 13 (Baby S., 1947), Negro male, was born after 7 mo.' gestation. He went steadily downhill for no obvious reason, and died on the 12th day of life despite transfusions and sulfonamides.

Autopsy (9 days post mortem, the body having been kept in a refrigeration compartment) showed: omphalitis due to cocci; kernicterus; focal subependymal encephalomalacia with meningeal infiltrate of microglia; rickets; hemosiderosis of liver and spleen; 5 cc. hemoperitoneum; minimal fibrinous peritonitis; superficial gastric erosions and ulcerative enterocolitis with pneumatosis of the ascending colon.

Microscopic examination showed small erosions, without inflammatory reaction, in the gastric lamina propria. The small intestine showed an area of plasma cell exudate underlying a serosal fibrin deposit, an area of deep necrosis with lymphocytic reaction, and congestion of submucosal vessels with diffuse lymphocyte and plasma cell infiltrate. The colon showed multiple irregular mucosal ulcers, mostly in the distal end. The cysts were submucosal, containing serum, lymphocytes, monocytes and giant cells. A few small vessels (veins or lymphatics) in the mesenteric LN contained fibrin and degenerated lymphocytes.

Discussion

Poor general condition was a common feature in these patients; four of the infants were premature, and all the infants were clinically considered small, weak or athreptic. Cases 5 and 8 had abnormally low resistance to staphylococcal infection. There was a high incidence of abnormalities related to the gastrointestinal tract that either were congenital or developed in the neonatal period: hypertrophic pyloric stenosis (two instances),

rectal prolapse, reduplication of small intestine, pancreatic fibrosis, intrahepatic biliary obstruction, and malrotation of the colon. Extraintestinal disease was absent in only one patient (Case 11); of the other 12 patients, 10 had respiratory infections—otitis, mastoiditis and pneumonia—with one instance (Case 5) of lung abscess and empyema. Case 7 had peritonitis and liver failure, Case 13 had omphalitis, kernicterus and hemosiderosis, and Case 12 had, in addition to otitis and mastoiditis, fulminating septicemia. These findings parallel those listed in table 1.

The gastrointestinal complaints in these patients were similar to those reported elsewhere. Seven patients had diarrhea, 6 had vomiting, 5 had abdominal distention and 3 had gross blood in the stools.

The distribution of the lesions also parallels that previously reported. It was: colon 9, ileum 6, jejunum 2, colon and ileum 3, jejunum and ileum 1. The mesenteric lymph nodes were examined microscopically in several cases, but showed only atrophy.

Concerning the pathogenesis, the close relation between a small mucosal defect and the immediately distal pneumatosis in Case 7 is of interest. Case 10, where the pneumatosis underlay a superficial ulcer not covered by membrane, also suggests a mucosal defect as the point of entrance of the gas. In Case 8 the pneumatosis occurred near a thrombosed vessel in the cecum, and in Case 11 the pneumatosis was most marked where the ulceration was most marked.

Microscopically, the cysts were predominantly submucosal, containing air and occasionally protein-like fluid. There was no clearcut evidence that bacteria had entered the intestinal wall along with the gas, but there was evidence, in Case 6 and perhaps also in Case 7, of secondary infection of ruptured cysts. The numerous submucosal giant cells in Case 8 were considered to be reaction to the cysts; only the subserosal giant cells were attributed to the presence of catgut sutures.

This is the first series of infants in which the duration of the process can be estimated. In Case 2 the cysts were demonstrable by RG 9 days before death, in the same region found involved at autopsy. In Case 7, nine days antemortem, and in Case 8, 20 days antemortem, no pneumatosis was seen on RGs or at extensive laparotomy. In Case 11 the pneumatosis may have been present 20 days antemortem but not observed during pyloromyotomy.

Conclusion

The chief interest of this admittedly unimportant condition is the theoretic questions it raises. No case reported in the literature holds pneumatosis responsible for death of a child or infant. However, a full explanation of the pathogenesis of pneumatosis might give useful information about the disturbed intestinal physiology in infantile diarrhea, although a causal relationship cannot be definitely established. It can only be stated that diarrhea is the most frequent complaint of infants found at autopsy to have pneumatosis. The exact conditions needed to produce gas cysts, and the frequency of pneumatosis in infants who survive diarrhea, are not known. Present concepts of disturbed electrolyte balance need no discussion here; however, little is known of the nature of the derangements severe enough to cause death despite the best modern therapy. To recapitulate, pneumatosis in adults occurs most often in intestine whose function is already disturbed, by ulceration proximal to, or by compromised circulation in, the affected segment. Experimental and clinical evidence indicate that low-grade nonspecific infection is either prerequisite or adjuvant in keeping the gas confined within the intestinal wall instead of its being rapidly absorbed

by the mesenteric lymphatics, and microscopic mucosal defects are frequent. In adults, removal of the peptic ulcer or a band constricting a loop of intestine results in disappearance of the pneumatosis. If such change in physiology could be understood and produced deliberately in infants suffering from diarrhea, the cause of both pneumatosis and electrolyte imbalance might be removed.

Summary

Thirteen cases of pneumatosis intestinalis are reported, 12 of them in infants between 12 days and 12 months of age, one in a boy of 6 years. Review of these cases and of 32 reported cases falling within the pediatric age range discloses that the disease occurs most frequently in patients whose general condition is poor, that it is very often associated with congenital or acquired disease of the intestine, and that respiratory disease, usually infectious, frequently co-exists. The presence of pneumatosis in pediatric patients has so far been discovered only at autopsy, but clinical diagnosis, with the aid of the typical roentgenologic findings, is feasible and may be accomplished when the disease is more widely known.

The clinical picture and roentgenographic findings in adults are reviewed. The most acceptable theories concerning the pathogenesis are discussed, with their possible relation to infantile diarrhea.

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SPANISH ABSTRACT

Pneumatosis Intestinalis; Revisión de la Literatura y Reporte de 13 Casos

Se reportan 13 casos de pneumatosis intestinalis, de ellos 12 en infantes de 12 días a 12 meses de edad, y un caso en un niño de 6 años. La revisión de estos, así como de otros 32 casos referidos en distintas comunicaciones (todos estos pacientes menores de 14 años de edad) revela que la enfermedad ocurre más frecuentemente en sujetos cuya condición general es pobre, o que sufren de otra enfermedad congénita o adquerida del intestino, o de una afección respiratoria, generalmente de tipo infecciosos. Hasta el presente, la pneumatosis en enfermos pediátricos ha sido descubierto solamente en la autopsia, pero el diagnóstico clínico, con la ayuda de los rayos X (que muestran un cuadro típico), es posible y se hará con más frecuencia cuando la enfermedad sea mejor conocida.

Se revisan el cuadro clínico y los signos radiológicos en los pacientes adultos. Se discuten las teorías más aceptables concernientes a la patogénesis. Se espera que elucidando la causa de esta enfermedad se obtenga también información en el mecanismo de producción de la diarrea infantil a la cual comunmente va asociada.

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OUTBREAK OF AEROBACTER INFECTIONS ON INFANTS' WARDS

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A STUDY on the control of airborne infection by means of triethylene glycol vapor was carried out on the infants' wards of the Harriet Lane Home, Johns Hopkins Hospital, between Nov. 1, 1945, and May 1, 1946. Certain aspects of this study are described elsewhere.[1-3] The original plan was to follow carefully the introduction on the ward and subsequent spread of the various types of pneumococcus and hemolytic streptococcus. However, late in December 1945, there began to appear in the nasopharyngeal cultures of patients an encapsulated gram-negative rod which showed pronounced capsular swelling in the presence of antipneumococcic rabbit sera types 23 and 32, thus making it easy to identify in cultures. This organism spread subsequently in more or less epidemic fashion on the wards, giving rise to both clinical and subclinical infections. It was recovered not only from the nasopharynx but also from the environment on many occasions over a period of several months.

Since clinical infections due to encapsulated gram-negative rods of the Klebsiella-Friedländer-Aerogenes group are apparently being recognized with increased frequency,[4,5] these organisms are perhaps of greater interest to clinicians than formerly. The present report deals with a self-limited outbreak of infection due to such a pathogen and sheds some light both on the classification of members of this group and on the manner in which they spread from person to person.

PLAN OF CROSS-INFECTION STUDY

The study referred to here has already been described in some detail.[1] Briefly, 2 identical infants' wards of the Harriet Lane Home were employed in the study, 1 being kept as a "control" ward, while in the "test" ward triethylene glycol was vaporized. Patients were admitted alternately to the 2 wards, thus insuring a population similar with respect to age, incidence of bacterial disease, etc. A fairly rigorous aseptic technic was followed when caring for the infants in order to minimize the opportunity for direct transfer of organisms under study.

Two nasopharyngeal cultures were obtained from each infant on admission, 2 each week thereafter and 1 at the time of discharge. Appropriate cultures were taken from all patients developing secondary infections. In addition each patient was examined on alternate days for evidence of infection. Nose and throat cultures were taken routinely once a week on the doctors, nurses and ward attendants, and from January through March on the visitors. Semiweekly bacteriologic surveys of the environment were carried out, including cultures of the table tops, chairs, bedding, nurses' hands and uniforms. Pneumococci and streptococci were identified with particular care. From early January on, the specific gram-negative rod which gave rise to this report was carefully sought in all

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cultures, whether of patients, personnel or environment. This organism will henceforth be referred to as Aerobacter "Carter" after the patient from whose nasopharynx it was first cultured.

Bacteriologic Data

Morphology and Cultural Characters: The bacillus under discussion is, when freshly isolated from a patient, a short, nonmotile, pleomorphic, gram-negative encapsulated rod, which grows readily on solid as well as in liquid media. Enrichment of the medium with blood or glucose hastens growth but is by no means essential. The colonies produced on agar are large, glistening, gray or yellowish gray, and very mucoid. The colony on an agar plate, when touched with a platinum loop, will "spin a thread." A 24 hour growth

TABLE 1

Biochemical Reactions of 2 Strains of Aerobacter

	Aerobacter "Carter"	Aerobacter "Hettinger"
Lactose	AG	AG
Glucose	AG	AG
Maltose	AG	AG
Mannite	AG	AG
Sucrose	AG	AG
Xylose	AG	AG
Rhammose	AG	AG
Arabinose	0	AG
Indole	0	0
Gelatin	No liq.	No liq.
Litmus milk	Coag.	Coag.
Hemolysis of rabbit's red cells	0	0
Hemolysis of sheep's red cells	0	0
Motility	0	0

in broth often exhibits a slimy gray pellicle, particularly around the edges of the tube. There is abundant ropy sediment in the old broth cultures. The capsule is very large and is readily seen on hanging drop preparations to which a loopful of methylene blue has been added. The capsule often appears to be unequally "thick" around the different parts of a single organism, appearing larger at the "ends" than along the "sides." When specific antiserum is added to such a preparation, the capsules swell enormously, so that the bacillus appears as a small blue dot or oval body within a transparent globe.

Table 1 sums up the biochemical reactions of Aerobacter "Carter" and of another closely related organism, Aerobacter "Hettinger," isolated at about the same time from a patient with pyelitis. Both organisms produce acid and large amounts of gas within a few hours in glucose, lactose, maltose, sucrose, mannite, xylose and rhamnose. Both coagulate milk and fail to liquefy gelatin. There is no hemolysis of rabbit's or sheep's red cells. Streptomycin was found to inhibit the growth of Aerobacter "Carter" in a concentration of 7.8 u./cc. of culture medium, thus placing the sensitivity of the organism midway between that of S. typhosa and H. influenzae.

Mouse Virulence: Mice succumb to intraperitoneal injection of amounts greater than 0.5 cc. of a 10^{-3} dilution of a 24 hour broth culture (approximately 250,000 organisms), so that this organism is of relatively low virulence for mice.

Antigenic Structure: Until recent years no satisfactory division of the Klebsiella-Aerobacter group had been made on the basis of antigenic structure. It was then shown by Avery, Heidelberger and Goebel,[6] and also by Edwards[7] and Julianelle,[8] that the immunologic reactions of this group, similar to those of the pneumococci, depend on the presence of two factors: a capsular polysaccharide and a somatic nucleoprotein. For classification it is necessary to prepare serum by the injection of smooth, capsulated bacilli containing both capsular and somatic antigens.

The authors attempted to elucidate the relationship of Aerobacter "Carter" to other capsulated pathogenic bacteria.

Sera against 3 different strains of Aerobacter* were prepared in the laboratory. Rabbits were immunized with a vaccine consisting in each case of an 18 hr. broth culture of the organism, attenuated, but not completely killed, by the addition of a few drops of formalin. The serum of each rabbit was tested for presence of antibody before immunization and was found negative. The vaccine was administered intravenously 3 times a week for 4 wk. starting with a dose of 0.1 cc. and gradually increasing to 2.0 cc. The animals were bled from the heart 2 to 3 wk. after the last injection of vaccine. Specific sera against types 23 and 32 Pneumococcus were prepared by Lederle Laboratories and supplied by the Office of the Surgeon General.

"Absorbed sera" were prepared as follows: Pneumococci were grown for 36 hr. in beef infusion broth containing 1% dextrose. Gram-negative rods were grown on blood agar plates and washed off with saline. The broth and saline suspensions, respectively, were centrifuged and the supernatant discarded. The serum to be absorbed was added in a volume equivalent to half the volume of packed organisms. After thorough shaking the mixture was allowed to stand for 48 hr. in the icebox, then centrifuged. The clear supernatant serum was the "absorbed serum" used in the capsular swelling reactions. All capsular swelling preparations were set up in duplicate on different days, and each was examined by 2 observers.

The results of the capsular swelling tests are set forth in tables 2 and 3. It can be seen that Aerobacter "Carter" showed capsular swelling with its crude homologous antiserum, even when that antiserum was remarkably dilute (1/8172). It showed capsular swelling, but to a lower dilution, in the presence of anti-"Carter" serum absorbed with pneumococcus type 32, and to a still lesser degree in the presence of antipneumococcic serum type 32. On the other hand, type 32 pneumococcus (strain "Weldon") showed capsular swelling with its homologous antiserum out to a high dilution, but only to a low dilution in the presence of crude anti-"Carter" serum. Absorbing the type 32 serum with Aerobacter "Carter" lowered its capsular swelling titer for pneumococcus type 32 only slightly. Aerobacter "Carter" showed capsular swelling in the presence of unabsorbed

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* For the bacterial strains the authors are indebted to the following:

Miss H. D. Zepp for Aerobacter "Hettinger," isolated at Sydenham Hospital, Baltimore, from the urine of a patient with pyelitis.

Dr. Dwain Walcher for Aerobacter "Yale #7," isolated at the New Haven Hospital, New Haven, Conn., from the stool of an infant with epidemic diarrhea.

Miss Annabel Walter, Bureau of Laboratories, Department of Health, New York, for Pneumococcus type 23 "McMechen" and type 32 "Lanform."

Pneumococcus type 32 "Weldon" was recovered from a patient on the wards at the time of this study.

"Sydenham #5" refers to a strain of Aerobacter isolated in January 1945 from a Negro infant admitted to Sydenham Hospital, Baltimore, with pertussis complicated by bronchopneumonia. It showed capsular swelling in the presence of type 23 antipneumococcic serum but not with type 32 antiserum. The organism itself had been lost, but a specific rabbit serum prepared at that time was made available by Miss H. D. Zepp.

antipneumococcic serum type 23 diluted 1/128, but the relationship between these organisms was not studied in detail. In the presence of sera against pneumococci of types other than 23 and 32, Aerobacter "Carter" displayed no capsular swelling.

Pneumococcus type 23 (strain "McMechen") and pneumococcus type 32 (strains "Lanform" and "Weldon") showed capsular swelling in the presence of anti-"Carter"

TABLE 2

RESULTS OF CAPSULAR SWELLING TESTS WITH AEROBACTER "CARTER"

	Dilutions of Serum*														
	1	2	4	8	16	32	64	128	256	512	1024	2048	4086	8172	16344
Anti-"Carter" serum: unabsorbed	+	+	+	+	+	+	+	+	+	+	+	+	+	+	−
Absorbed with "Carter"	−	−	−	−	−	−									
Absorbed with Pneumo. type 32	+	+	+	+	+	+	+	+	+	−					
Antipneumococcic serum type 32: Unabsorbed	+	+	+	+	−	−	−								
Absorbed with "Carter"	−														
Antipneumococcic serum type 23: Unabsorbed	+	+	+	+	+	+	+	+	−	−					

* Expressed in reciprocal of dilution.

serum, but here again the relationship was not studied in detail.

It seems clear then that Aerobacter "Carter" and type 32 pneumococcus have a common capsular antigen, but that each of these organisms contains, in addition to the common antigen, one or more other antigens. It is also clear that Aerobacter "Carter" and type 23 pneumococcus share a common capsular antigen.

In table 4 can be seen the results of capsular swelling reactions set up between several strains of Aerobacter, H. influenzae, Friedländer's bacillus and meningococcus. Aerobacter "Carter" appears to share one or more common capsular antigens with each of two other strains of encapsulated gram-negative bacilli ("Hettinger" and "Sydenham #5") as well as with types 23 and 32 pneumococcus. One other recently isolated gram-negative rod ("Sydenham #5") shares a capsular antigen with type 32 but not with

type 23 pneumococcus. Aerobacter "Carter" seems not to share any capsular antigen with H. influenzae, meningococcus or Friedländer's bacillus.

An attempt was made to find out whether strains "Hettinger" and "Yale #7" might by chance be related to some type of pneumococcus. No such relationship could be demonstrated.

The fact that definite interrelationships were demonstrated between strains "Carter,"

TABLE 3

RESULTS OF CAPSULAR SWELLING TESTS WITH PNEUMOCOCCUS TYPE 32 ("WELDON")

	Dilutions of Serum*														
	1	2	4	8	16	32	64	128	256	512	1024	2048	4086	8172	16344
Anti-"Carter" serum: Unabsorbed	+	+	+	±	−										
Absorbed with "Carter"	−	−	−												
Absorbed with Pneumo. type 32	−														
Antipneumococcic serum type 32: Unabsorbed	+	+	+	+	+	+	+	+	±	−					
Absorbed with "Carter"	+	+	+	+	+	+	+	−	−						

* Expressed in reciprocal of dilution.

"Hettinger," "Yale #7" and "Sydenham #5" is interesting, since two of these strains were recovered originally from the upper respiratory tract, one from the urine and one from a stool culture.

Such sharing of capsular antigens by apparently unrelated bacteria is not surprising, since heterogenetic specificity has frequently been described.[6, 9-12] Such interrelationships are probably much more frequent than we realize, since, in the course of routine bacteriologic investigation, it is not usual to set up encapsulated members of the coli-aerogenes group in antipneumococcic sera to look for capsular swelling.

CLINICAL OBSERVATIONS

Incidence of Gram-Negative Rods in Nasopharyngeal Cultures

Of the 361 patients included in this study, 174 were found on one or more occasions to harbor gram-negative rods in their nasopharynx. Of these 174, 50 were found to be Aerobacter "Carter." This organism was first identified on Dec. 20, 1945. Within the next

four months, it was recovered from the nasopharyngeal cultures of 50 different infants ranging in age from 7 days to 2 years. In 23 patients, it was recovered on one occasion only, in 9 on 2 occasions, in 11 on 3 occasions, in 2 on 4 occasions. Cultures were positive in individual patients as often as 5, 6, 7, 8 and even 9 times in a row.

Effect of Chemotherapy on Incidence

The first patient from whom Aerobacter "Carter" was isolated was an infant who had been receiving large doses of both penicillin and sulfadiazine for a period of six weeks. Subsequently, however, the likelihood of a patient's picking up this organism seemed independent of whether or not he was receiving chemotherapy. Nineteen patients who had received no antibiotic within 5 days or more picked up the organism; 15 patients had been receiving penicillin for periods of 2 to 17 days before the first positive culture; 4 patients had received both penicillin and sulfadiazine together for 3 to 17 days previously.

TABLE 4

SEROLOGIC RELATIONSHIPS OF AEROBACTER "CARTER" AND OTHER ENCAPSULATED BACTERIA

Antiserum:	Organism						
	"Carter"	"Hettinger"	"Yale #7"	Pneumo 23 "McMechen"	Pneumo 32 "Lanform"	Pneumo 32 "Weldon"	Friedländer Type B
Aerobacter "Carter"	8172*	2	Aggl	+	+	4	0
Aerobacter "Hettinger"	0	128	Aggl	–	–	0	0
Aerobacter "Sydenham #5"	4	1	0	–	–	4	0
H. influenzae, Type A	0	0	0	–	–	0	0
H. influenzae, Type B	0	0	0	–	–	0	0
Friedländer's bacillus, Type A	0	0	0	–	–	0	0
Friedländer's bacillus, Type B	0	0	0	–	–	0	+
Meningococcus Group I	0	–	–	–	–	–	–
Meningococcus Group II-a	0	–	–	–	–	–	–
Meningococcus Group IV	0	–	–	–	–	–	–

* Expressed in reciprocal of dilution.

Symptomatology

The evaluation of the symptoms produced by this organism was quite difficult. In five patients there is reason to believe that the acquisition of this organism may have been responsible for recognizable symptoms.

Patient 1 was a 1 month old Negro infant admitted for treatment of congenital syphilis. Nine days after entering the hospital he developed mild diarrhea consisting of 5 to 7 watery stools a day. This was not accompanied by fever, dehydration or other constitutional manifestations. He recovered after 10 days without any treatment other than restriction of his caloric intake. Four nasopharyngeal cultures during a nine day period and one stool culture were positive for Aerobacter "Carter."

Patient 2, an 8 month old infant admitted for study of a pulmonary lesion due either to tuberculosis or lipoid pneumonia, was afebrile for several days before her temperature spiked to 39.6° C.; at the same time Aerobacter "Carter" was found in her nasopharynx. She remained feverish for three days and her cultures remained positive for seven days.

Patient 3, a 1 year old child with a hemangioma, developed irritability, profuse nasal discharge, and marked swelling and redness of the tonsils coincidental with cultures positive for Aerobacter "Carter."

Patient 4, a 6 week old infant, was admitted with severe bronchopneumonia and atelectasis. He improved markedly at first, only to develop fever and diarrhea 7 days later at the time Aerobacter was first cultured from his nasopharynx. Three nasopharyngeal cultures during that period were positive.

Patient 5, an 11 month old boy with tetralogy of Fallot, developed pharyngitis and catarrhal otitis media one day after his nasopharyngeal culture first showed Aerobacter "Carter." Five positive cultures were obtained from him over a 14 day period.

Three patients showed a brief febrile episode at the time their cultures first became positive for Aerobacter. There were no accompanying symptoms, such as pharyngitis or diarrhea, and it was impossible to be sure whether it was a coincidence.

On another group of patients, four in all, an elevation of temperature coincided with the appearance of the encapsulated rod, but also with some other event which would provide adequate explanation for fever: an operative procedure, onset of rubeola, etc.

The remaining patients from whom Aerobacter "Carter" was recovered on culture showed no symptoms whatsoever attributable to a fresh bacterial infection at the time of their initial acquisition of this organism.

Epidemiologic Observations

From the time of the first positive culture for Aerobacter "Carter" (obtained Dec. 20, 1945), the incidence of positive nasopharyngeal cultures rose very slowly until early in March, when the increase became more rapid, the peak incidence of infection being reached in April. Thereafter the incidence of positive cultures fell off rapidly, and, without any special control measures being instituted, the organism disappeared entirely.

Coincidental with the large number of positive cultures obtained from patients, several members of the nursing staff also were found on one or two occasions to harbor Aerobacter "Carter" in their weekly nasopharyngeal cultures as follows:

March 12	ward maid	1
18	nurses	2
25	nurses	2
April 1	nurses	17
	attendant	1
	ward technician	1
2	nurses	10
	attendant	1

Aerobacter "Carter" was not recovered at any time from the members of the medical staff, who, however, on numerous occasions harbored other unidentified types of gram-negative rod, as did also the nurses and attendants. Aerobacter "Carter" was recovered on one occasion each from 14 of the adult Sunday visitors. One of these cultures was positive in February, one in March and 12 on April 14 toward the end of the peak incidence of infection among the patients. All these visitors had previously shown negative cultures, suggesting that they acquired the organism while visiting on the wards. Little is known of the incidence of encapsulated gram-negative rods in the nasopharynx of healthy individuals. Bloomfield,[13] in 1921, made careful studies on 85 normal adults and found five

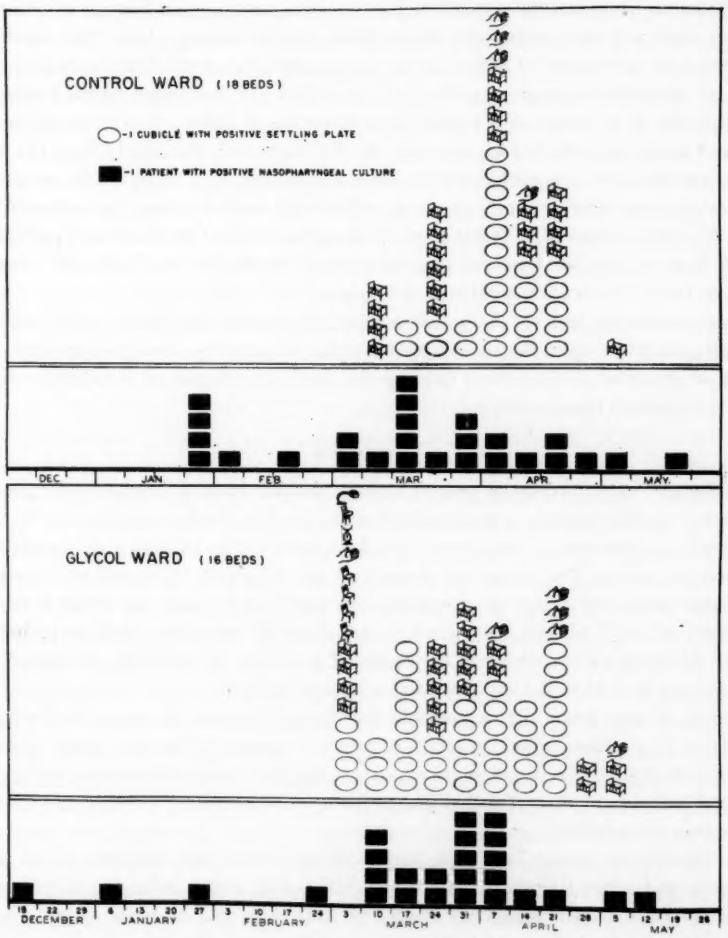

CHART 1. Graphic representation. Occurrence of Aerobacter "Carter" carriers week by week on both glycol and control wards; also, contamination of environment, as indicated by different symbols, in terms of positive cultures obtained from bedside settling plates, from cribs, hands, bedside chairs, scales, high chair and lamp.

of them to be harboring organisms which he termed Friedländer's bacillus; since typing of such organisms was not then possible, the organisms were identified by means of cultural characteristics which would place them, according to present definition, in the Klebsiella-Aerobacter group.

It is of interest that the ward environment also became heavily contaminated, as shown in chart 1. The contamination of the air was studied in two ways, (1) by bubbling measured amounts of ward air through nutrient broth, aliquots of the broth then being plated out to facilitate colony counts, and (2) by placing open pour plates on each bedside table and allowing the organisms from the air to settle upon them. From both bubbler samples and settling plates many colonies of Aerobacter were recovered. Chart 1 shows graphically the relationship between the number of patients with positive nasopharyngeal cultures on a given date, and the number of cubicles with positive settling plates. The widespread occurrence of Aerobacter "Carter" in the environment was confirmed by swab cultures taken on numerous occasions. On March 7, for instance, it was found on the hands, feet, face and hair of a patient with a positive nasopharyngeal culture, as well as on his shirt, the sheet of his bed, the bedside table top, the bed lamp and the cubicle floor. On March 9, the organism was recovered by culture from a highchair, baby scale, a table upon which clean masks were piled, a nurse's gown, as well as from several tables, chairs, beds and the floors of three cubicles in different parts of the glycol ward. On March 30, swabbing of 5 or 6 beds in each ward yielded positive cultures. Aerobacter was found on numerous occasions on the hands of both patients and nurses.

As can readily be seen on the accompanying chart, triethylene glycol vapor exerted no demonstrable effect upon the incidence of Aerobacter infection. On the glycolized ward, 25 out of a total of 183 patients picked up the organism, whereas on the control ward 25 out of 178 patients became infected.

Discussion

Aerobacter "Carter" is, by its general biologic characteristics, a member of the group of organisms variously known as Klebsiella, Aerobacter, Friedländer's bacillus and B. mucosus capsulatus. Members of this group have long been known to have a very general distribution in nature. They occur on floors,[14, 15] in the soil,[16, 17] as well as in water.[18] Introduced usually by means of a contaminated water supply, they can produce so-called "ropiness" of tea,[18] of milk and cream,[19] and of maple syrup,[20] as well as undesirable "slimy" fermentations occurring in the course of a number of industrial processes,[21] such as the tanning of leather and the manufacture of paper pulp.

Bacteria of this group are responsible for disease processes in many warm-blooded animals, such as guinea pigs,[22] mares,[23] mice[24] and moose.[25] They are closely related to the genus Erwinia,[26] the members of which are frequent plant pathogens, causing galls, wilts and soft rots of many familiar plants. They are also often present in the intestines of flies, bees and beetles.[27]

Man likewise is subject to a wide variety of infections with members of the genus Aerobacter and genus Klebsiella. Bamforth[28] collected 81 strains belonging to this group, all isolated from human beings. Of these, 19 were recovered from the nasopharynx, 9 from sputum, 32 from feces, 16 from urine and 5 from miscellaneous sources such as the conjunctiva or cervix. Perkins,[29] in the course of 286 consecutive autopsies, came across organisms of this group in 79 cases; at one time or another they were isolated from almost

all organs in the body. Obrinsky and associates,[30] in a recent article, review the literature, particularly with respect to the occurrence of Friedländer-aerogenes infections in children.

Encapsulated gram-negative rods have been known to be responsible not only for illnesses of individuals, but also on occasion for epizoötics[24, 31] as well as for epidemic spread of disease in humans: diarrhea in an obstetric nursery,[32] diarrhea in a children's home,[33] respiratory disease among prisoners of war.[34] In the course of investigating an epidemic of respiratory disease which occurred from March to December 1941, among raw-cotton workers, Caminita and associates[35, 36] found the low grade raw cotton, as well as the cotton mill dust, to be heavily contaminated with an encapsulated gram-negative rod; the authors suggested that repeated attacks of illness due to these organisms might be the cause of the chronic respiratory disease of raw-cotton workers known as byssinosis.

The wide distribution of members of the Aerobacter-Klebsiella group in nature and the relative frequency with which these pathogens are implicated in disease are not without importance for the clinician. In the first place, clinical bacteriologists sometimes classify an encapsulated rod in the genus Aerobacter if it is isolated from air, water, stool or urine, whereas if found in the human respiratory tract or meninges, it is called Klebsiella. From the evidence quoted above, such a differentiation seems unfounded and even undesirable, since it tends to suggest that a particular strain has a predilection for a particular portal of entry. The present authors incline to the view expressed by many who have studied the problem of classification[7, 30, 37-39] that the members of the genus Aerobacter and genus Klebsiella should be considered as a single group.

The mode of spread of infection with Aerobacter "Carter" is not so clear as the spread of hemolytic streptococci and pneumococci described in other reports in this series.[1-3] Direct patient-to-patient transmission was almost completely excluded by the use of separate cubicles for each patient. Indirect contact by means of hands and fomites was certainly rare, owing to the careful medical aseptic technic enforced. Healthy adult carriers (i.e., nurses, attendants) seem unlikely as major sources of infection. Bloomfield[13] studied this point with particular care: he placed asymptomatic carriers of Friedländer's bacillus in close contact with noncarriers, and found no spread from healthy carrier to contact. Moreover, as shown in other reports on the Harriet Lane Home study, the path of infection in the case of pneumococci and hemolytic streptococci appeared to lead from patient to nurse or attendant, rather than the reverse. There remain as probable sources of infection the air and the dust. As shown in chart 1, the entire ward environment (air, dust, furniture, bedclothes, etc.) became heavily contaminated and remained so for a period of two months. Each patient was thus surrounded by a heavily infected atmosphere. The source of contamination of the atmosphere itself is a matter for conjecture. At least two patients who harbored Aerobacter "Carter" in their nasopharynx had a purulent nasal discharge, and the importance of a purulent discharge from nose, ear or skin in the dispersion of pathogens is clearly shown by studies already referred to on the spread of pneumococci and streptococci. At least one infant also had positive stool cultures and diarrhea, and under such conditions a considerable number of organisms is probably dispersed into the environment. In support of the view that dispersion occurred mainly from respiratory discharges is the fact that the outbreak was self-limited, ceasing abruptly with the onset of warm weather and the disappearance of respiratory disease from the community.

Since Aerobacter "Carter," itself of low pathogenicity, contains nevertheless antigens

common to several other organisms, it is interesting to speculate upon the effect produced by it upon the immunity of some 50 infants admitted to the Harriet Lane wards in the course of this study. Many of them must have developed antibodies to types 23 and 32 pneumococcus without direct exposure to these organisms. There are undoubtedly innumerable other such "short cuts" in the acquisition of immunity. It is obvious also that under appropriate conditions these organisms might act as sensitizing agents, rendering an individual hypersensitive to substances to which he has never been directly exposed.

Summary

In the course of a study on the control of airborne infection by means of triethylene glycol vapor, there occurred an outbreak of infection due to a previously undescribed encapsulated gram-negative rod.

The morphologic and cultural characteristics of this organism place it in the Aerobacter-Klebsiella group. It was readily identified by its ability to show capsular swelling in the presence of sera against types 23 and 32 pneumococcus. Serologic studies showed it to be antigenically related to certain other strains of encapsulated, gram-negative rods, but not to Friedländer's bacillus, H. influenzae, meningococcus or pneumococcus other than the types mentioned above.

Some of the patients who acquired the organism developed mild respiratory infections or diarrhea, accompanied by fever, but most remained asymptomatic.

The organism spread in epidemic fashion on both glycolized and control wards and was recovered from the nasopharynx of 50 patients out of a total of 361 included in the study, also from numerous nurses, attendants and visitors. In the course of the epidemic the ward environment (air, dust, furniture, bedclothes, etc.) became heavily contaminated. It is postulated that patients with profuse nasal discharge and diarrhea may have served to infect the ward environment, which in turn served as the source of infection to other patients.

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SPANISH ABSTRACT

Brote Epidemico Debido al Aerobacter en una Sala de Niños

Un brote de infecciones debidas a un bacilo encapsulado, Gram-negativo no descripto previamente, ocurrió en el curso de un estudio sobre el control de infecciones transmitidas por el aire por medio del vapor de glicol trietileno. Las caracteristicas morfológicas y culturales de este organismo lo colocan dentro del grupo de las Aerobacter-Klebsiellas. El germen fué rapidamente identificado mediante su abilidad para mostrar edema de la capsula a los sueros de pneumococos tipo 23 y 32. Las pruebas serológicas demostraron relaciones antigénicas con ciertas clases de bacilos encapsulados Gram-negativos, pero no con el bacilo de Friedlander, el bacilo hemófilo influenzae, meningococos y otros pneumococos que los anteriormente citados. Algunos de los pacientes contaminados con el organismo en questión sufrieron de diarrea o presentaron un cuadro de un proceso infeccioso discreto de las vías respiratorias. El brote se desarolló en forma de epidemia tanto en las salas tratadas con glicol como en las no tratadas a los efectos de control del experimento. El organismo fué recuperado o cultivado de la nasofaringe de 50 pacientes de un total de 361 casos investigados, así como de varias enfermeras, auxiliares y visitantes. En el transcurso de la epidemia el ambiente de la sala (aire, polvo, mobilario, ropa de las camas, etc.) fué intensamente contaminado. Se postula que los enfermos con secreciones nasales profusas y diarreas hayan contaminado el ambiente de la sala y éste a su vez se transformó en la fuente de infección para otros enfermos.

1430 Tulane Avenue

SAFETY APPARATUS FOR INTRAVENOUS THERAPY

By JOHN P. FLETCHER, M.A., M.D.
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THE administration of potent drugs and fluids intravenously often requires much more accurate control than is possible with the usual drip method.

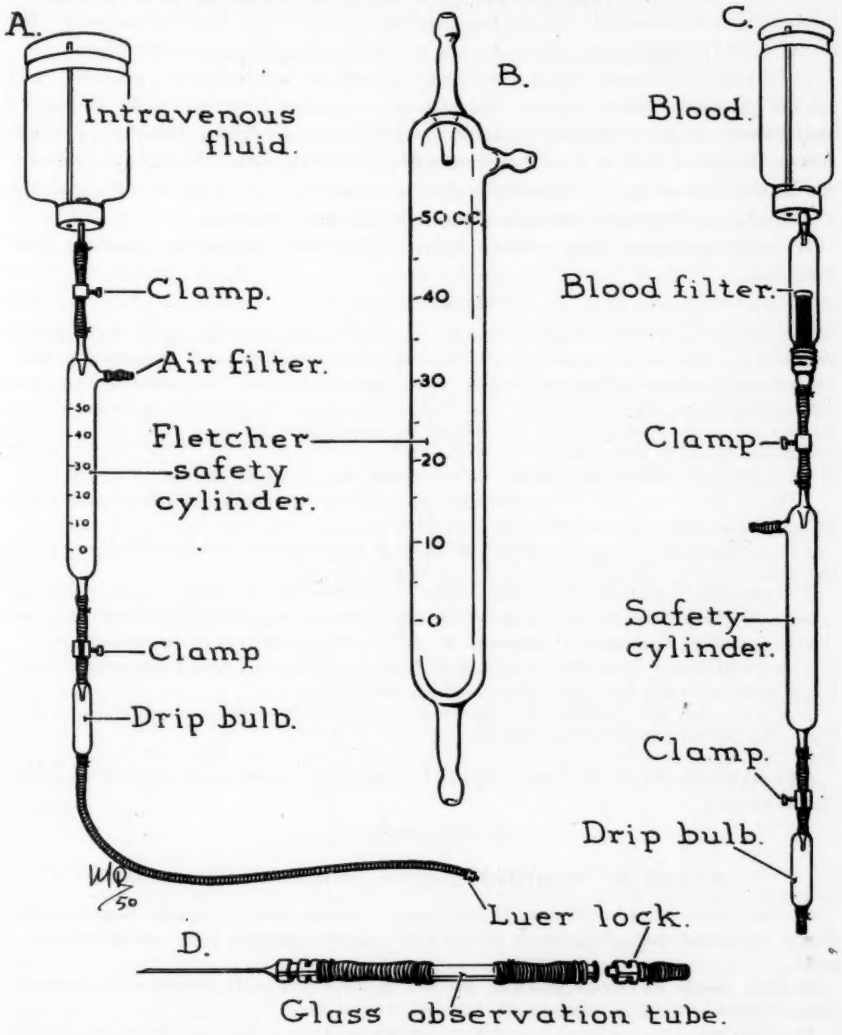

CHART 1

From the Hospital for Sick Children and the Department of Paediatrics, University of Toronto, Toronto.

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This is particularly applicable in the case of small infants for the following reasons:

1. Due to change in position of the infant or relaxation of spasm in the vein receiving the fluid, there may be large variations in the rate of flow, resulting in under- or over-hydration. In the latter case, excessive administration of water or specific electrolytes may cause death in an acutely ill patient.

2. Rates of flow of 10 to 15 cc. per hour are often desirable. It has been found to be impractical to measure a 15 cc. per hour outflow from a 500 cc. flask. Consequently, the nurses must frequently slow down the rate of flow so that stoppages often occur.

3. Certain intravenous medication—notably sodium sulphadiazine, penicillin and calcium gluconate—when injected into standard intravenous tubing, results in massive intermittent dosage. It is more desirable to give continuous intravenous therapy, maintaining the blood level at a uniform concentration. Crystallization of sodium sulphadiazine in the intravenous tubing has been observed when the drug is injected intermittently. This results in a stoppage of the intravenous and irritation of the vein.

A simple graduated safety cylinder (chart 1) has been designed to overcome these difficulties.

Method of Use

The safety cylinder (B) is inserted between the large container for intravenous fluids and the drip bulb (as shown in A) or between the blood filter and the drip bulb (as shown in C). While administering intravenous fluids or blood the clamp immediately above the safety cylinder is kept closed except when filling the cylinder. The fluid is delivered to the patient from the cylinder via the drip bulb. Soluble drugs are injected through the tubing below the upper clamp into the safety cylinder and mixed by gentle shaking of the cylinder.

The advantages of using this method for intravenous therapy are as follows:

1. In case of a "run away," a maximum of only the 70 cc. contained in the cylinder could flood the infant. This amount is most unlikely to cause death.

2. It is simple for the nurse to measure accurately in the cylinder the amount of fluid given each half hour. Therefore it is easy to keep the flow at the prescribed rate.

3. Continuous medication with soluble drugs is simple when the drug is injected into the cylinder. The drug is thereby diluted into 70 cc. Crystallization does not occur and irritation of the vein is reduced to a minimum. A constant rate of flow of the drug is easily maintained.

4. In transfusion, a blood filter is incorporated into the system (C). Accuracy in rate of transfusion has been found to be of great value in exchange transfusion.

5. If a very slow rate is desired, the intravenous solution may be heparinized with heparin 1 mg./100 cc. and the intravenous run intermittently.

This apparatus may be obtained from J. F. Hartz Co., Toronto, or from Fisher Burpe Co., Winnipeg.

Spanish Abstract

Aparato de "Seguridad" para la Terapeutica Intravenosa

El autor estima que la administración intravenosa de drogas potentes y fluidos, frecuentemente requiere un control exacto imposible de obtener en los aparatos corrientes de los metodos de gota a gota.

El autor describe un cilindro graduado, diseñado en forma a evitar la administración demasiado rápida o demasiado lenta de soluciones de electrolitos o de sangre.

El metodo para su uso es simple y es graficamente demostrado en el diagrama. El cilindro graduado de "seguridad" es llenado con la solución de electrolitos o sangre contenida en el frasco corriente. Se cierra el "clamp" superior y entonces el fluido o sangre contenido en el cilindro de seguridad es administrado al paciente a traves del cuenta gotas.

180 St. Clair Avenue East

PLASMA AND ERYTHROCYTE CHEMISTRY OF THE NORMAL INFANT FROM BIRTH TO TWO YEARS OF AGE

By RICHARD R. OVERMAN, Ph.D., JAMES N. ETTELDORF, M.D., ANNE C. BASS, A.B., AND G. B. HORN, M.D.

(With the technical assistance of Louise Spearman, B.S.)

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THE increasing interest in and attention to clinical problems relating to water and electrolyte metabolism in pediatrics and other branches of medicine emphasizes anew the need for more precise knowledge of normal physiologic growth and differentiation. That the processes of growth and development are attended by a greater variability or even instability of chemical anatomy than appears in the adult is attested to by almost every research into conditions arising in the neonatal period and in infancy. Our questionable proclivity to derive far-reaching interpretations of water and ionic shifts after making a series of fundamental assumptions regarding normal chemical anatomy of the adult is, thus, even less justifiable in pediatric research. Numerous contemporary comments on results of therapeutic regimes in infant diarrhea, in pernicious vomiting, in diabetic acidosis, in "hypoadrenalism," and in renal insufficiency in children, as well as interpretations of therapeutic trials of ACTH and cortisone in a variety of neonatal and infantile diseases would gain in validity if the disease-altered chemical anatomy were compared to adequate normal values for the particular age group studied.

It is the purpose of this paper to present direct measurements of the following variables in a sufficient number of normal babies and children in serial age groups to confer statistical validity on the results: Plasma and erythrocyte concentrations of Na, K, and Cl; plasma concentrations of Ca, NPN, protein and Co_2; plasma pH and venous hematocrit.

Previous reports of normal chemical values in the neonatal period, in infancy and during childhood include those of Bruch and McCune,[1] Marples and Lippard,[2] McCance and Young,[3] Miller,[4] Dodd and Rappaport,[5] Lucas et al.,[6] Young and Hallum,[7] Hoffman et al.,[8] Hoag and Kiser,[9] Erickson et al.[10] and Macy.[11] Many of these reports deal with but few variables and none report erythrocyte values in children under 5 years of age. Only the work of Bruch and McCune and Marples and Lippard deals with values for serial age groups.

CLINICAL MATERIAL AND METHODS

Determinations of the previously listed variables were made on venous blood in a total of 188 normal infants from birth to 2 yr. of age as well as in 19 normal women at term and in 19 arterial samples from the umbilical cord before delivery of the placenta.

Sodium and potassium concentrations were measured in plasma and packed erythrocytes using a Model 52-A Perkin-Elmer internal standard flame photometer. The samples were prepared according to the method of Overman and Davis[12] except that 93.5 parts/million lithium solution was used to make the original dilution in order that the filtrate, following protein precipitation, would contain

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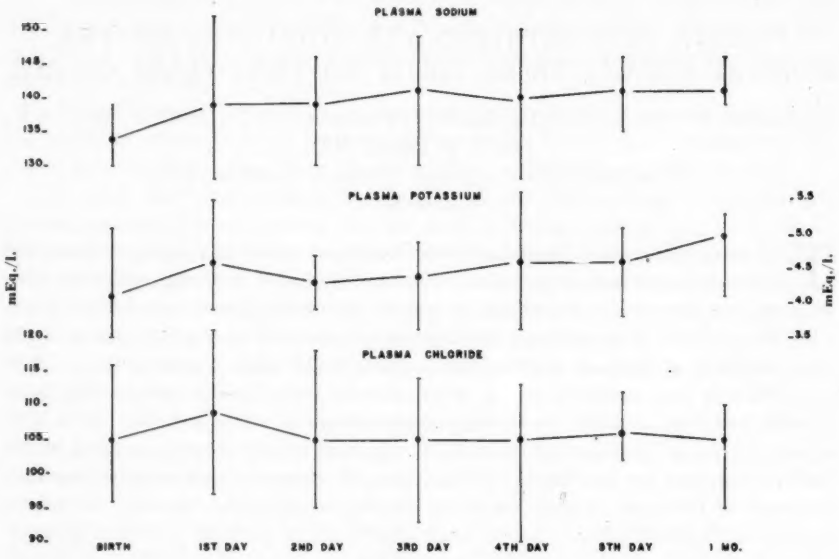

CHART 1

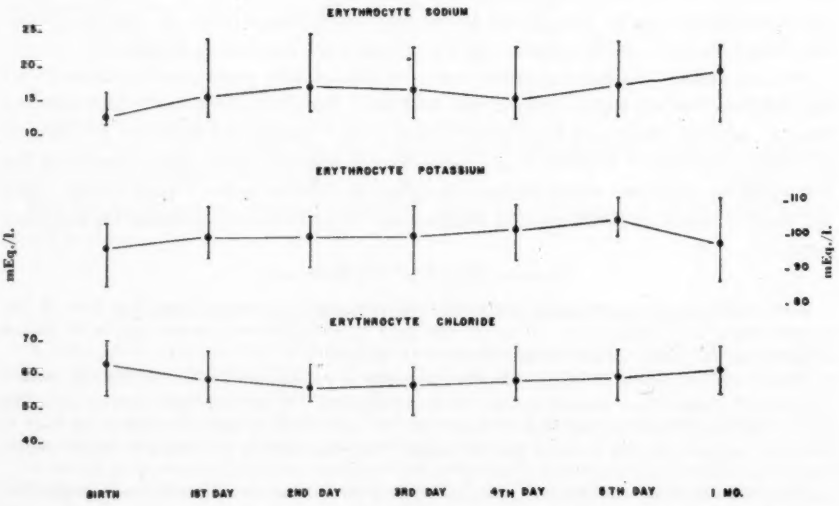

CHART 2

70 parts/million lithium as an internal standard. All further dilutions were made using 70 parts/million lithium solution, and all standard solutions were made up to contain that concentration of lithium ion. Calibration curves for each maximum standard were made at the time of each determination.

Chloride was measured by a modification of the Volhard method.[33] Blood samples for analysis were withdrawn into oiled, heparinized syringes and delivered under oil to prevent the "chloride shift."

Plasma calcium levels were determined by the Clark-Collip modification of the Kramer-Tisdall potassium permanganate titration method.[34] The micro-Kjeldhal method of Folin-Wu was used to determine plasma NPN.[35]

Plasma protein concentration was determined by the copper sulfate method of Phillips et al.[36]

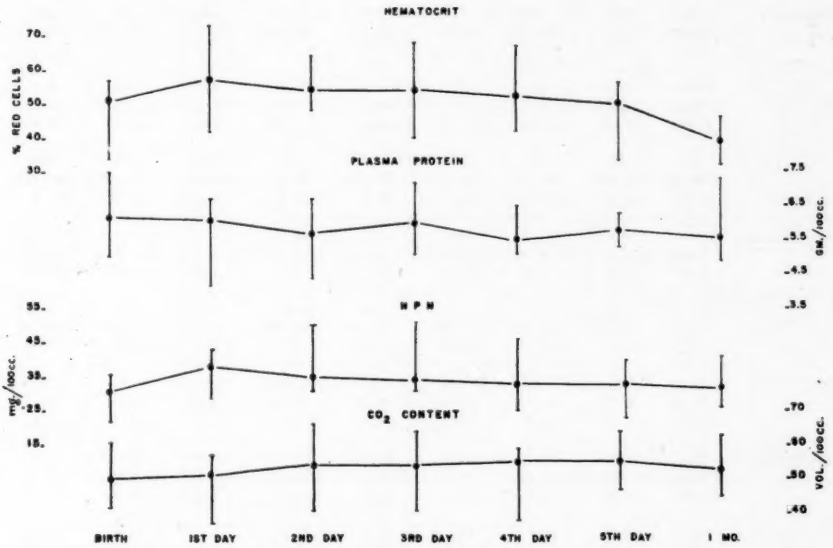

CHART 3

with confirmatory checks at intervals by the Weichselbaum micro-method.[37] The Van Slyke manometric method was employed to measure CO_2 content.[38] Hematocrit values were established using Wintrobe tubes which were centrifuged 45 min. at 2300 g. Plasma pH was measured using a Beckman glass electrode pH meter.

RESULTS

The results of this study are presented in tabular and graphic form. Charts 1, 2 and 3 are line graphs representing daily alterations in the measured variables from birth through the fifth day of life. For comparison, the one month averages are shown as well. The vertical lines on these graphs indicate the ranges. Each point represents the average of from 8 to 22 cases (table 1). Although each point represents an average of a different group of children, the points have been connected in order more clearly to indicate trends. In the bar graphs, the first bar indicates the average of all babies from birth through the fifth day of life (104 cases). The last bar indicates the average of 50 adult values determined previously using the same methods reported here.

Plasma Ions: The average plasma Na concentration in the blood of the newborn infant was identical with that of maternal blood and umbilical artery blood (table 1). Within the first 24 hours (chart 1) plasma Na rose to 140 mEq./l. at which level it remained throughout the period of time measured (tables 1 and 2). The constancy of the plasma

TABLE 1

Variable		Normal mothers	Cord blood	Babies, newborn	Babies, 24 hr.	Babies, 48 hr.	Babies, 72 hr.	Babies, 96 hr.	Babies, 102 hr.
No. Cases		19	19	17	22	20	17	20	8
Plasma Na	Ave.	134	134	134	140	139	141	140	141
	Range	126–129	126–141	130–139	128–152	130–146	130–149	128–150	135–146
	S.D.	±3.4	±4.2	±2.8	±4.4	±4.7	±5.0	±6.3	±3.6
K	Ave.	3.8	5.0	4.1	4.6	4.3	4.4	4.6	4.6
	Range	3.2–4.7	3.8–7.2	3.5–5.1	3.9–5.5	3.9–4.7	3.6–4.9	3.6–5.6	3.8–5.1
	S.D.	±0.41	±1.01	±0.49	±0.40	±0.30	±0.37	±0.74	±0.42
Cl	Ave.	102	109	105	109	105	105	105	106
	Range	94–108	99–117	96–116	97–121	95–118	93–114	89–113	102–112
	S.D.	±3.1	±6.3	±4.9	±8.2	±5.8	±6.6	±5.9	±3.8
Red Cell Na	Ave.	13.4	13.0	12.5	15.5	16.9	16.2	14.9	17.0
	Range	10.4–19.1	10.8–16.1	11.3–16.1	12.6–23.9	13.2–24.5	12.2–22.6	12.1–22.6	12.6–23.4
	S.D.	±2.4	±1.8	±1.3	±2.8	±3.0	±2.6	±2.5	±3.7
K	Ave.	97	98	97	100	100	100	102	105
	Range	89–104	91–107	86–104	94–109	91–106	89–109	93–109	100–111
	S.D.	±4.3	±4.5	±6.2	±4.0	±4.1	±4.7	±3.8	±4.1
Cl	Ave.	61	63	63	59	56	57	58	59
	Range	49–71	53–76	54–70	52–67	52–63	48–62	52–68	52–67
	S.D.	±7.2	±7.0	±6.8	±4.5	±4.1	±3.8	±4.3	±5.6
Hematocrit	Ave.	38.8	50.7	51.1	56.6	53.9	53.7	52.0	50.1
	Range	30.3–50.8	37.2–64.0	34.3–57.1	41.9–73.1	48.0–63.6	40.0–68.1	42.0–66.5	33.3–66.5
	S.D.	±5.9	±5.4	±5.4	±7.2	±4.6	±8.3	±5.5	±7.2
CO_2 Content	Ave.	49.5	50.9	49.7	50.5	53.9	53.7	54.9	55.0
	Range	37.9–53.4	38.5–58.2	42.4–6.0	37.4–56.9	40.9–66.5	41.4–63.8	38.0–58.5	47.4–64.1
	S.D.	±5.3	±5.1	±7.5	±4.6	±6.0	±6.4	±5.2	±5.7
pH	Ave.	7.47	7.39	7.38	7.43	7.43	7.42	7.42	7.39
	Range	7.42–7.61	6.79–7.51	7.15–7.61	7.32–7.56	7.31–7.59	7.24–7.59	7.30–7.52	7.29–7.54
	S.D.	±0.44	±0.16	±0.12	±0.62	±0.63	±0.81	±0.73	±0.24
NPN	Ave.	25	32	31	38	35	34	33	33
	Range	20–31	26–38	22–36	29–43	29–50	29–51	25–46	23–40
	S.D.	±3.4	±3.6	±4.1	±4.8	±5.7	±5.8	±5.7	±5.0
Plasma Protein	Ave.	6.8	6.3	6.2	6.1	5.7	6.0	5.5	5.8
	Range	5.9–7.5	5.5–7.7	5.1–7.5	4.2–6.7	4.4–5.7	5.1–7.2	5.1–6.5	5.3–6.3
	S.D.	±0.35	±0.62	±0.57	±0.70	±0.52	±0.51	±0.48	±0.35

Na concentration in the healthy adult is well known and this phenomenon apparently extends to within the first day of life.

The plasma K concentration was high in umbilical artery plasma, the average being 5.0 mEq./l. with a range of from 3.8 to 7.2 mEq./l. This finding is a confirmation of that reported by Earle, Farber and Alexander[19] (table 1). The newborn infant had a plasma K concentration slightly higher than maternal levels (table 1). The plasma K

TABLE 2

Variable		Babies, 1 mo.	Babies, 2–4 mo.	Babies, 5–7 mo.	Babies, 8–10 mo.	Babies, 11–15 mo.	Babies, 19–26 mo.
No. cases		14	16	16	12	15	11
Plasma Na	Ave.	141	142	141	140	139	141
	Range	139–146	137–153	137–155	138–142	135–144	138–145
	S.D.	±2.2	±4.3	±4.2	±2.4	±2.6	±2.1
K	Ave.	5.0	4.7	4.5	4.5	4.6	4.2
	Range	4.1–5.3	4.0–5.4	3.8–5.3	4.1–4.9	3.8–5.4	3.5–4.7
	S.D.	±0.56	±0.40	±0.48	±0.26	±0.47	±0.39
Cl	Ave.	105	106	105	103	104	104
	Range	95–110	104–109	96–120	101–106	102–107	101–108
	S.D.	±6.0	±1.6	±5.0	±1.9	±2.4	±2.1
Ca	Ave.	5.4	5.3	5.4	5.3	5.4	5.1
	Range	5.2–5.7	4.8–5.6	5.2–5.6	5.1–5.8	4.7–5.6	4.9–5.3
	S.D.	±0.17	±0.24	±0.95	±0.23	±0.22	±0.22
Red Cell Na	Ave.	19.0	19.8	19.8	19.4	19.7	18.7
	Range	11.6–22.9	12.8–28.9	13.8–28.0	17.2–24.3	15.6–26.2	15.2–25.0
	S.D.	±3.6	±3.8	±3.4	±3.9	±2.7	±2.6
K	Ave.	98	100	99	105	107	101
	Range	87–111	83–112	91–107	96–117	100–115	91–115
	S.D.	±6.5	±7.3	±5.6	±6.0	±5.0	±6.4
Cl	Ave.	61	65	62	65	65	68
	Range	54–68	60–72	57–72	56–72	53–76	61–74
	S.D.	±4.3	±4.3	±4.1	±5.2	±7.2	±4.5
Hematocrit	Ave.	39.4	35.2	34.4	34.6	33.7	36.5
	Range	32.1–45.5	30.2–40.4	30.8–39.1	30.7–41.0	30.6–38.2	31.1–39.3
	S.D.	±3.6	±2.2	±2.5	±3.1	±2.7	±2.2
CO_2 Content	Ave.	52.6	52.9	55.1	54.0	53.8	55.2
	Range	45.1–62.9	43.2–60.1	46.1–63.4	41.6–64.0	39.6–60.0	48.3–60.3
	S.D.	±7.7	±5.9	±4.7	±5.5	±5.3	±3.8
NPN	Ave.	32	28	32	—	30	29
	Range	26–41	27–29	25–41		28–32	26–31
	S.D.	±5.4	±1.0	±4.8		±1.5	±1.5
Plasma Protein	Ave.	5.6	6.0	6.3	6.5	6.7	6.8
	Range	4.9–7.3	5.1–6.9	5.7–7.0	5.7–7.3	6.3–7.0	5.9–7.3
	S.D.	±0.73	±0.24	±0.37	±0.52	±0.26	±0.52

concentration rises gradually from 4.1 mEq./l. at birth to 5.0 at one month and then decreases again to 4.2mEq./l. at 2 years (tables 1 and 2, charts 1 and 4).

Chloride concentrations in the plasma were quite constant, being 105 mEq./l. at

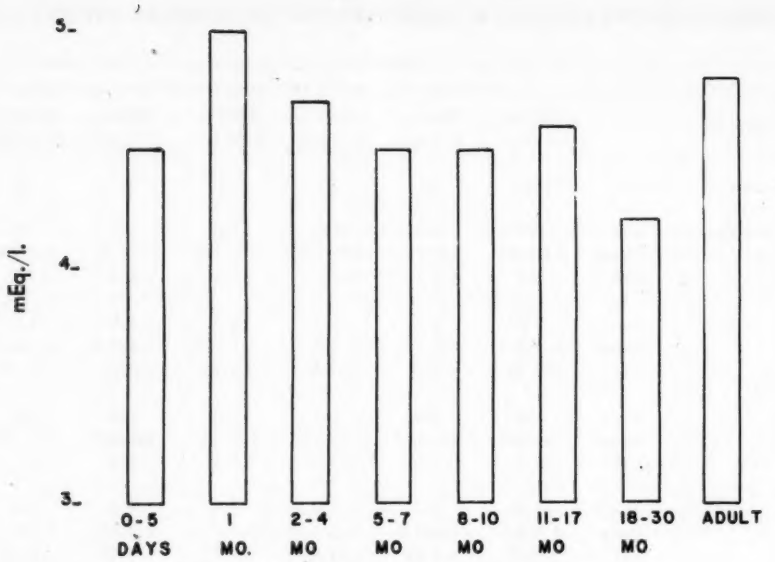

CHART 4. Normal plasma potassium concentrations.

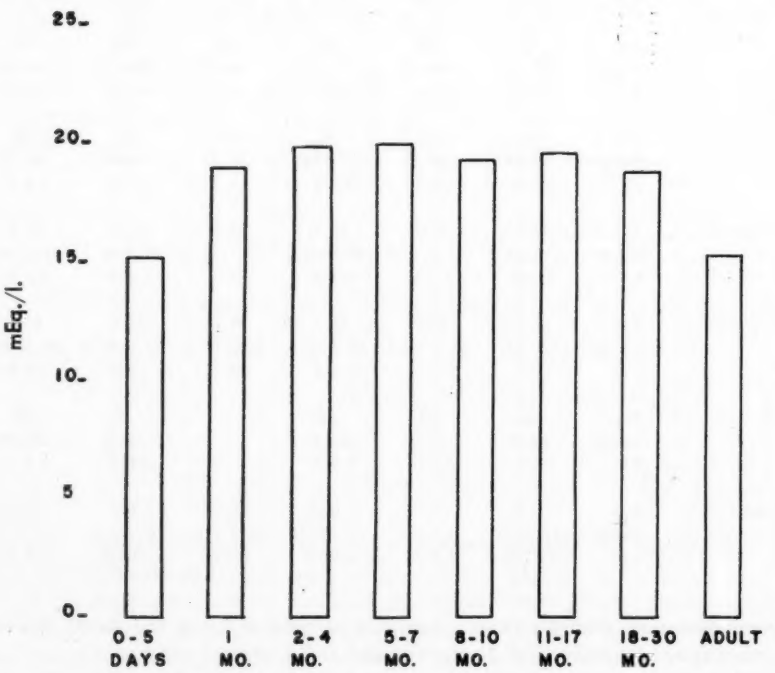

CHART 5. Normal erythrocyte sodium concentrations.

birth and, with the exception of the 24 hour sample, remaining 105 to 106 mEq./l. until one year when a slight reduction to 103 to 104 mEq./l. occurs (tables 1 and 2, chart 1).

Plasma Ca concentrations were determined only in the six groups of babies studied between the first month and second year (84 cases). Again, a remarkable constancy was found, the average being 5.3 or 5.4 mEq./l. up to and including the 15 month old

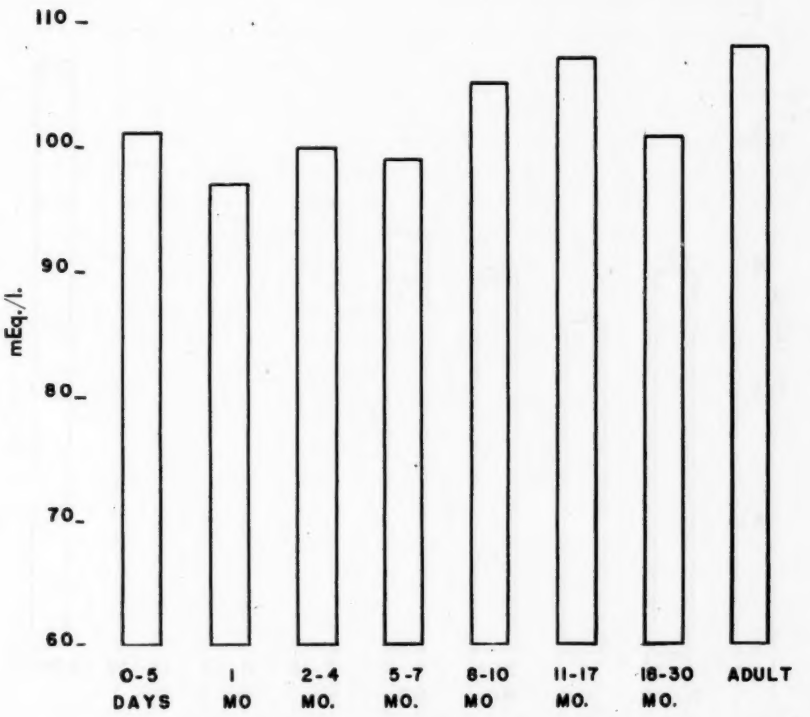

CHART 6. Normal erythrocyte potassium concentrations.

group. A slight reduction to an average of 5.1 mEq./l. was noted in the 19 to 26 month old series (table 2).

Erythrocyte Ions: The erythrocyte Na concentration, although it was the same at birth as that of the normal adult (table 1), gradually increased until the average for all groups of babies between 2 and 17 months old was 20 mEq./l., approximately 25% higher than the adult normal (charts 2 and 5). With the exception of one baby whose red cell Na was 25 mEq./l., the cell Na level tended to be lower at the age of 2 years.

Variations in erythrocyte K were smaller. The trend was toward higher cell K levels in the older babies; the greatest increase (about 8%) occurred between the ages of 1 and 17 months (charts 2 and 6).

At birth the red cell chloride level was higher than the adult normal average of 54 mEq./l. in both the mothers (61 mEq./l.) and the babies (63 mEq./1.). It had decreased slightly by the following day but began immediately to rise again and continued to increase throughout all the age groups measured until it was 68 mEq./l. at 2 years of age, 20% higher than the adult normal level (tables 1 and 2).

Venous Plasma NPN: With the exception of a slight rise to 38 mg./100 cc. at 24 hours

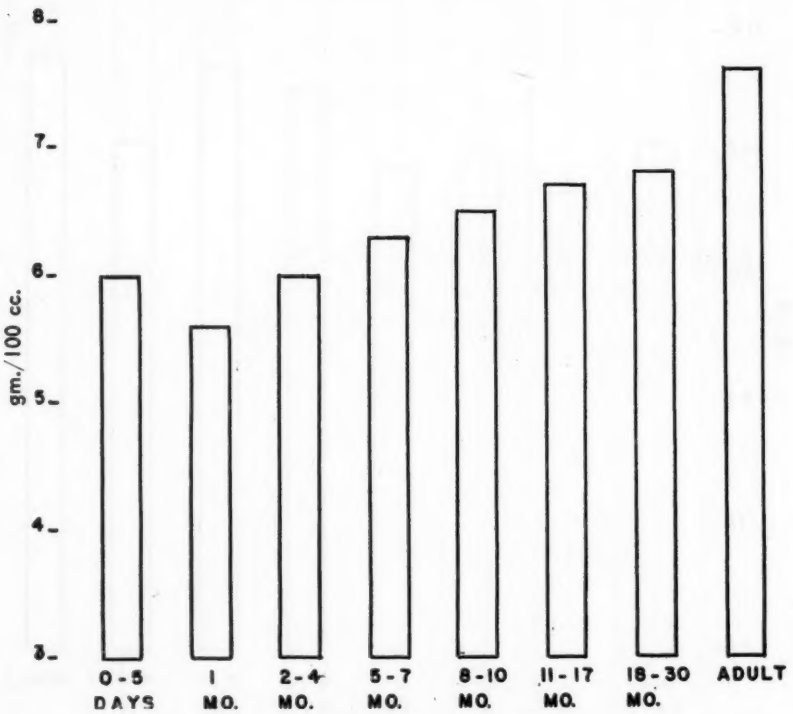

CHART 7. Normal plasma protein concentrations.

after birth, the plasma NPN remains at adult normal levels throughout the period studied (tables 1 and 2, chart 3).

Carbon Dioxide Content: The CO_2 content of the venous plasma showed a slight rise from an average birth value of 49.7 to 55.0 volumes per cent by the fifth day and remained unchanged thereafter.

Venous Plasma pH: The plasma pH remained constant at 7.4 from 1 through 5 days of life (table 1). The average for the group sampled at birth was 7.3 with a range of 7.2 to 7.6; this difference is probably not significant since the standard deviation is $\pm .12$.

Plasma Protein Concentration: At birth the plasma protein concentration of the babies averaged 6.2 gm./100 cc. compared to 6.8 gm./100 cc. for the mothers (table 1), and by the second day of life it had decreased to 5.7 gm./100 cc. (chart 3). Between 2 and 7 months the protein concentration began to increase again and continued to do so for all

age groups measured until it reached 6.8 gm./100 cc. at 2 years of age (chart 7 and table 2).

Venous Hematocrit: From 51.1% at the time of birth the venous hematocrit increased to 56.6% during the first 24 hours (chart 3). Subsequently the hematocrit continued to decrease until the age of 2 to 4 months when it was 35.2% (chart 8). During the first year of life it remained at this low level (normal adult range 42 to 50%); by the second year it had increased to 36.5% (table 2).

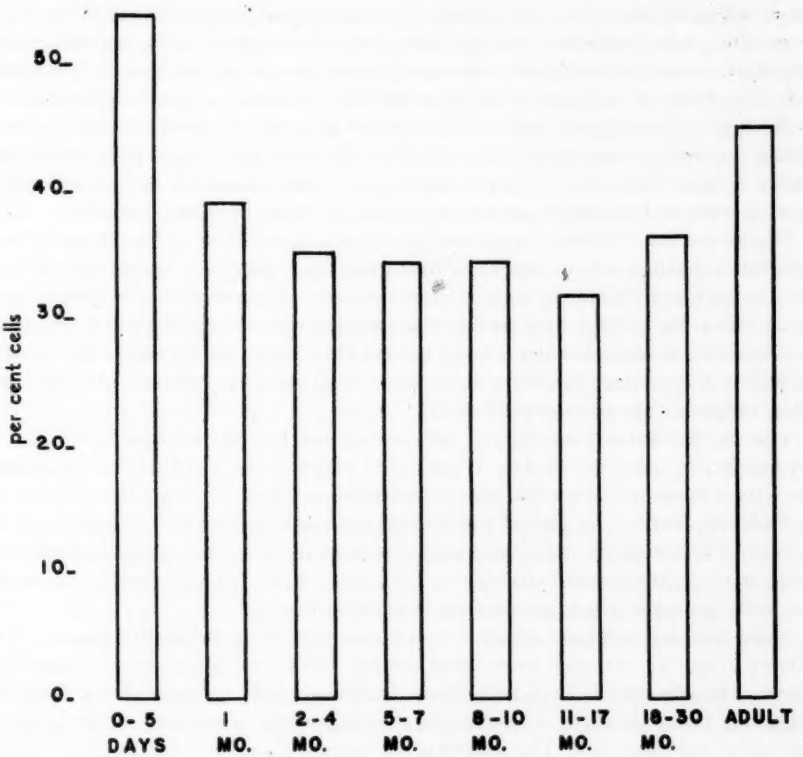

CHART 8. Normal hematocrit values.

DISCUSSION

Water and electrolyte metabolism during normal neonatal life has apparently escaped extensive study. Perhaps the best extant review of the literature appears in Clement A. Smith's *Physiology of the Newborn Infant*[20] in connection with considerations of renal and adrenal functions. In the section on renal physiology he states, "Few studies from large numbers of subjects are available as sources for accurate standards of the normal plasma electrolytes in the newborn infant." A search of the literature since the appearance of this work fails to reveal any major additional sources of such data. While the emphasis of such studies as were done prior to 1945 was on kidney function, more recent interest in

normal standards is centered about adrenal cortical function in the newborn infant and about diseases common to the neonatal period in which water and/or electrolyte depletion is a salient feature.

The inclusion of electrolyte analysis of packed erythrocytes in the present study was made in an effort to reveal at least directional changes regarding cellular ions. The authors realize that the erythrocyte is scarcely a representative body cell, still it provides cellular material which can be secured essentially free of extracellular fluid.* Further, the ionic pattern of the intracellular fluid of primate erythrocytes, unlike that of the carnivores, varies independently of the extracellular ionic pattern and is apparently well protected from direct renal regulation. Perhaps alterations in erythrocytic ionic patterns do not constitute a simple reflection of tissue cellular ionic change, yet they may be of value in indicating trends of water and/or ion movement between intra- and extracellular fluids.

As might be anticipated, more striking alterations in the measured variables occurred within the first 24 hour period of extrauterine life than during any other comparable period of time. This critical interval might well be more intensively studied with reference to water and electrolyte metabolism as well as other physiologic variables.

During the first 24 hours, increments in plasma concentrations of Na, K and Cl and in hematocrit values indicate water loss from the entire extracellular compartment as well as from the plasma. While the range in plasma protein concentrations tended to be reduced at 24 hours, the average total protein concentration was not materially different from that measured at birth. If water is being lost from the plasma during this period, protein as well is disappearing, possibly to be utilized in the metabolic process during the time when exogenous nutriment is withheld.

That the lost water is not entering cells is suggested by the concomitant 24% rise in erythrocyte Na and a 3% increase in red cell K (charts 2 and 3). After the first month, the almost linear rise in plasma protein concentration (chart 7) might be explained in at least three ways: 1. A gradual and continual plasma dehydration. If this occurs at all it must be of but minor importance since it is not reflected by rising plasma ionic concentrations. 2. An increased efficiency in the production of plasma protein. 3. A steady rise in the sparing of plasma protein from metabolic destruction.

A continual but nonlinear reduction in hematocrit occurs up to the fifth month of life (charts 3 and 8), the most pronounced portion (30%) of which occurs in the first month. The reduction in hematocrit occurs concomitant with a rise in plasma K values suggesting that erythrocytic destruction is at least partially responsible for the reduced number of red blood cells. The somewhat low average plasma K value for the 18 to 30 month old babies is inexplicable.

The alterations in erythrocytic K concentrations during normal development from birth to 2 years are not large and are probably not of significance. There is a general tendency toward an increase in erythrocytic K storage from one month (average 98 mEq./l.) to 17 months (average 107 mEq./l.). The normal adult average is closer to the latter. However, until the seventh month of life, red cell K concentrations are below 100 mEq./l. which is below the normal range for adults (103 to 116 mEq./l.). On the other hand, the erythrocyte Na is normal or slightly below normal at birth, rises almost con-

* The packed erythrocytic samples were not washed prior to analysis, thus a small but constant amount of plasma was included in the cellular aliquot. The authors' experience with primate erythrocytes indicates that between 4 and 5% of the packed sample is plasma and that this is constant.

tinuously until the seventh month and is still significantly above normal adult values at 30 months. The radiosodium space of the newborn infant has been reported by Flexner, et al.[21] to be 43.5% of the body weight. This figure is 1.7 times as large as the usually accepted adult normal value of 25% of the body weight. Such values have been thought to represent an actual elevation in extracellular fluid volume in infants, but the discrepancy could likewise be explained on the basis of Na entrance into cells. The high erythrocytic Na values reported in this study emphasize this possibility.

Since but few serial determinations of normal values have been made previously, comparison of these figures with those of others is difficult—especially because this data must be compared with several different series obtained by different groups using varying methods.* Among the plasma determinations made, the greatest departure of this data from that previously reported is in the plasma K values. McCance and Young,[3] using an adaptation of Denes iodometric micro-method for K, reported that the average plasma K for 18 infants from 7 to 14 days old was 7.8 mEq./l. However, they simultaneously reported an adult (25 to 41 years) average normal value of 5.6 mEq./l. which is somewhat high when compared to that obtained by other methods, including flame photometric studies.

The implications of the present report are (1) that values for the chemical electrolyte anatomy of infant body fluids vary considerably from adult normal values, (2) that progressive alterations in these values occur from birth to 2 years of age and (3) that clinical interpretations based on a substitution of adult for infant values may well be in error both quantitatively and qualitatively. For instance, while it may be true that infants with diarrheal disease exhibit a higher intracellular Na concentration than the normal adult, the present data suggest that this may not be a chemical abnormality in this age group.

Summary

One hundred eighty-eight normal infants from birth to 2 years of age have been studied in a serial manner with regard to concentrations of Na, K, and Cl in plasma and erythrocytes; plasma Ca, NPN, protein and CO_2; plasma pH and venous hematocrit. These values have been compared with those of normal women at term and with other adult normal values. Nineteen samples of blood from the umbilical artery were likewise analyzed for comparison.

Serial and progressive alterations in the measured variables occurred and in several regards the normal infant body fluid chemical anatomy departs sharply from normal adult values.

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* Since the studies reported here were done, serial age group determinations of serum Ca, inorganic phosphate, K, sugar, urea, citric acid, total protein and phosphatase have been done in 235 newborn infants by Gittleman and Pincus[22] and were presented before the Society for Pediatric Research at the 1950 meeting at French Lick, Ind.

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SPANISH ABSTRACT

Análisis Quimico del Pasma y Eritrocitos en Niños Normales Desde el Nacimiento a los Dos Años de Edad

Un total de 88 niños normales entre el nacimiento y dos años de edad, fueron estudiados en una forma seriada con respecto a las concentraciones de sodio, potasio, y cloruro en los eritrocitos y en el plasma; y calcio, NPN, CO_2, proteina y pH en el plasma; y del hematocrito en sangre venosa. Estos valores fueron comparados a los obtenidos en pacientes normales con gestación a termino y otros valores normales de adultos. Asimismo, se análizarion 19 muestras de sangre obtenidas del cordon umbilical. En este estudio se observaron alteraciones seriadas y progresivas de las substancias medidas, indicando que los valores normales de la anatomia quimica del cuerpo del niño departe notablemente del adulto normal.

The University of Tennessee

VOLUMETRIC AND MICROSCOPIC PATTERN OF BONE MARROW IN NORMAL INFANTS AND CHILDREN

I. Volumetric Pattern

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SINCE the study published in 1937 by Kato,[1] there have been several reports of the sternal marrow findings in normal infants and children.[2-4] Ten years after Kato's report, Wolman[5] reviewed the literature relating to bone marrow examination in childhood; additional reports have been published recently.[6-8] None of these, however, deals with the volumetric and histologic data that can be obtained by the aspiration technics described and modified by Schleicher[9,10] and others.[11,12]

Osgood and Seamans[13] gave extensive consideration to the variables that should be controlled to arrive at normal values for bone marrow microscopic anatomy. They suggest that "The ideal . . . would permit removal of a large, representative, measured specimen in such form that accurate cell identification . . . may be obtained and all structural relationships could be observed. . . ."

Compared to the original direct smear technic, it is not agreed that the volumetric or other quantitative methods give additional reliable information.[11,12,14-16] However, the aspiration and volumetric technic employed in this study was adopted because preliminary observations indicated that one obtained information closely approximating, if not equaling, the ideal suggested by Osgood et al. In addition, since volumetric technics are being used routinely in some clinics for the study of adults' bone marrow, it would seem that in any event the determination of normal values for infants and children is desirable.

The purpose of this study, therefore, was (1) to ascertain the technical applicability of the volumetric method to be described to the study of bone marrow in infants and children, (2) to determine the influence of growth on the various data obtained, (3) to compare the data obtained from tibial, iliac and sternal aspiration and (4) to ascertain satisfactory sites for the study of bone marrow by this method at various pediatric age levels.

MATERIAL

Selection of Subjects: Seventy-two aspirations were performed on 50 normal infants and children. All individuals except the 17 newborn infants were patients in the Los Angeles Children's Hospital. For the most part they had been admitted for elective repair of congenital hernia or correction of strabismus. All elective cases were examined by pediatricians a few days prior to entry and again on the day of entry and were found to be in good health and free of evidence of acute or chronic infection.

Blood counts and urinalyses were done prior to surgery; if any distinct abnormalities were found, the patients were discharged without operation and hence were not included in this study. Table 1 lists the clinical diagnoses, peripheral blood findings and the volumetric data from all cases studied. Since urinalyses were all negative, they were not included in this table.

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TABLE 1

CLINICAL, PERIPHERAL BLOOD, TECHNICAL AND VOLUMETRIC DATA; 72 ASPIRATIONS, 50 NORMAL INFANTS AND CHILDREN

Subj. No.	Age	Admission Diagnosis	Date	Peripheral Blood							Bone Marrow							
				Hgb., gm./100 cc.	Leukocytes, thousands/mm³	Neutrophiles, %	Eosinophiles, %	Basophiles, %	Lymphocyte, %	Monocytes, %	Site of Aspiration†	Amount of Aspiration, cc.	Rate of Aspiration§	Fat, vol. %	Plasma, vol. %	ME, vol. %	Erythrocytes, vol. %	Particles, approx. no. and size
1	5 hr.	Normal newborn	2-49	—	—	—	—	—	—	—	T	2.0	+	0	40.0	4.0	56.0	0
2	5 hr.	Normal newborn	2-49	—	—	—	—	—	—	—	T	1.0	+	0	25.0	9.0	66.0	*
3	5 hr.	Normal newborn	3-49	—	—	—	—	—	—	—	T	1.0	+	0	32.0	11.0	57.0	0
4	5 hr.	Normal newborn	3-49	—	—	—	—	—	—	—	T	1.0	+	0	22.0	5.0	73.0	0
5	7 hr.	Normal newborn	3-49	—	—	—	—	—	—	—	T	0.75	+	0	29.0	7.5	63.5	0
6	9 hr.	Normal newborn	2-49	—	—	—	—	—	—	—	T	2.0	+	0	20.0	9.0	71.0	0
7	13 hr.	Normal newborn	2-49	—	—	—	—	—	—	—	T	1.0	+	0	34.0	9.0	57.0	*
8	15 hr.	Normal newborn	3-49	—	—	—	—	—	—	—	T	No Material Obtained						
9	18 hr.	Normal newborn	2-49	—	—	—	—	—	—	—	T	0.25	±	0	51.0	9.0	40.0	*
10	3 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	1.5	+	0	32.0	6.0	62.0	†
11	3 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	2.0	+	0	40.0	5.0	55.0	0
12	3 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	2.0	+	0	20.0	3.0	77.0	20 med.
13	4 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	1.0	+	0	30.0	5.0	65.0	†
14	4.5 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	1.5	+	0	29.0	3.0‡	71.0	0
15	4.5 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	2.0	+++	0	33.0	4.0	63.0	0
16	5 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	2.0	+	0	38.0	8.0	54.0	0
17	6 days	Normal newborn	3-49	—	—	—	—	—	—	—	T	3.0	+++	0	46.0	4.0	50.0	10 small
18	2 wk.	Meningocele	11-48	15.3	9.4	31	3	0	40	25	T	—	—	0.0	37.0	7.0	56.0	15 med.
19	2 mo.	Hernia	8-48	12.2	11.3	32	1	0	59	8	T	—	—	0.0	60.0	9.0	31.0	10 small
20	3 mo.	Hernia	12-48	10.0	7.[illegible]	28	10	1	54	7	T	4.0	+++	0.0	44.0	4.0	46.0	15–20
21	3 mo.	Hydrocele	10-48	11.3	17.3	22	2	0	65	11	T	3.0	+++	Tr.	55.0	5.0	40.0	†
22	4 mo.	Intussusception	8-48	12.7	12.3	60	5	0	34	5	T	3.0	+++	0.5	62.5	4.0	33.0	†
23	5 mo.	Hernia	10-49	10.8	6.8	29	4	0	61	6	T	3.0	+++	2.0	57.0	6.0	35.0	100 med.
24	8 mo.	Hernia	—	11.3	14.7	58	3	0	33	5	T	2.5	+++	2.5	55.0	4.0	38.5	40 med.
25	11 mo.	Hernia	12-48	10.5	7.2	23	1	1	67	8	T	—	—	2.0	40.0	4.0	54.0	0
26	13 mo.	Hernia	12-49	11.2	11.5	50	3	0	42	5	T	—	—	1.0	57.5	1.5	40.0	10–20 sm.
27	16 mo.	Hernia	11-48	13.6	13.4	57	2	0	33	8	T	3.0	+++	2.0	51.0	1.0	46.0	0
28	16 mo.	Phimosis	10-49	10.9	12.2	40	1	1	51	7	T	2.5	++	4.0	48.0	3.0	45.0	30 sm. & med.
29	19 mo.	Hernia	10-48	11.0	13.4	33	1	0	60	5	T	3.0	+++	1.5	50.0	1.5	47.0	20 large
											I	3.0	+++	0.5	49.0	3.5	47.0	0
30	19 mo.	Sebaceous cyst	12-48	13.0	7.4	32	3	0	59	6	T	—	—	5.0	45.0	5.0	45.0	15 med.
											I	—	—	1.0	46.0	7.0	46.0	15 med.
31	2 yr.	Hernia	11-48	13.6	5.9	34	6	1	53	6	T	4.0	+++	2.0	52.0	2.0	44.0	0
											I	3.0	+++	2.0	50.0	4.0	44.0	0

30	19 mo.	Sebaceous cyst	12-48	13.0	7.4	32	3	0	59	6	T I	— —	— —	5.0 1.0	45.0 46.0	5.0 7.0	45.0 46.0	15 med. 15 med.
31	2 yr.	Hernia	11-48	13.6	5.9	34	6	1	53	6	T I	4.0 3.0	+++ +++	2.0 2.0	52.0 50.0	2.0 4.0	44.0 44.0	0 0
32	2 yr.	Hemangioma	8-48	13.2	13.7	29	2	1	63	5	T I	3.0 3.0	+++ +++	1.5 0.5	52.0 50.0	2.5 5.0	44.0 44.5	0 0
33	2 yr.	Hernia	11-48	12.6	7.3	50	1	1	45	3	T I	4.0 1.5	+++ +	4.0 0.5	46.0 48.0	7.0 7.0	43.0 44.5	75 large 0
34	3 yr.	Hernia	11-48	12.1	9.9	42	6	0	43	4	T I	3.0 3.0	+++ +++	4.0 Tr.	46.0 45.0	2.5 4.0	47.5 50.0	0 0
35	3 yr.	Hydrocele	10-48	12.3	5.8	36	5	0	54	5	T I	5.0 2.0	+++ +++	3.0 1.0	51.0 51.0	1.0 3.5	45.0 44.5	0 10–20 sm.
36	4 yr.	Sacral anomaly	10-48	12.9	7.5	40	5	0	47	9	T I	3.0 2.0	+++ +++	6.0 1.5	55.0 51.0	1.0 4.0	44.0 43.5	0 0
37	4 yr.	Hernia	11-48	13.3	14.9	65	7	1	18	9	T I	3.0 2.0	+++ +	3.0 Tr.	49.0 49.0	1.0 5.0	47.0 46.0	0 0
38	4 yr.	Strabismus	10-49	13.6	5.3	46	0	0	48	6	T I	3.0 3.0	+++ +++	3.0 1.0	45.0 46.0	1.0 3.0	51.0 50.0	50 med. 25 sm.
39	4 yr.	Strabismus	12-48	12.1	8.3	43	3	0	51	3	T I	4.0 2.0	+++ +	1.0 1.0	49.0 48.0	1.5 5.0	48.0 46.0	Few min. 25 sm.
40	4 yr.	Strabismus	12-48	13.8	13.8	46	4	0	44	6	T I	4.0 3.0	+++ +++	4.0 1.0	49.0 50.0	0.5 4.0	46.0 44.0	0 0
41	5 yr.	Strabismus	11-48	13.6	6.8	47	6	0	40	3	S I	3.0 3.0	+++ +++	1.0 1.5	50.0 49.5	3.0 2.0	46.0 49.5	20 sm. & lg. 0
42	6 yr.	Hernia	8-48	12.9	10.8	50	1	0	43	6	S I	3.0 3.0	+++ +++	0.5 0.5	49.0 46.0	4.0 5.0	46.5 48.5	25 med. 0
43	6 yr.	Cataract, Enucleation	12-48	12.0	6.3	57	2	1	42	4	S I	— —	— —	0.5 2.0	51.0 48.0	4.0 5.0	44.5 45.0	† 10–15 lg.
44	7 yr.	Hypospadius	9-48	16.3	7.1	41	1	0	50	8	S I	3.0 2.0	+++ +	1.0 1.0	46.0 42.0	2.0 3.0	51.0 54.0	0 0
45	7 yr.	Esotropia	12-48	12.5	9.4	56	0	1	34	9	S I	5.0 2.0	+++ +++	Tr. 1.0	50.0 53.0	3.0 6.0	47.0 51.0	0 0
46	8 yr.	Phimosis	—	13.4	9.7	78	1	0	16	5	S I	2.5 5.0	+ +++	1.0 1.0	54.0 52.0	3.0 3.0	42.0 44.0	25 sm. 50 sm.
47	10 yr.	Phimosis	9-48	13.8	9.1	43	2	0	51	6	S I	3.0 3.0	+++ +++	0.5 0.5	51.0 53.0	2.0 1.0	46.5 45.5	0 0
48	11 yr.	Hypospadius	12-48	13.9	5.8	40	3	1	44	11	S I	3.0 5.0	+++ +++	3.0 2.0	38.0 38.0	7.0 4.0	52.0 56.0	30 sm. & lg. 30 sm. & lg.
49	11 yr.	Hernia	9-48	12.4	6.5	63	3	0	29	5	S I	3.0 3.0	+++ +++	1.0 0.5	51.0 51.5	5.0 2.0	43.0 46.0	0 0
50	11 yr.	CNS birth injury suspected. Past history of asthma	8-48	13.1	6.8	52	6	0	35	7	S I	3.0 3.0	+++ +++	1.5 1.0	49.0 47.0	5.0 4.0	44.5 46.0	0 0

* Insufficient material
† Clotted
‡ T, Tibia; I, Ilium; S, Sternum.
§ +, 1 cc. approx. 20 sec. or more; ++, slowly; +++, average, 2–5 cc. in approx. 3 to 10 sec., ++++, 5 cc. in approx. 3 sec. or less.

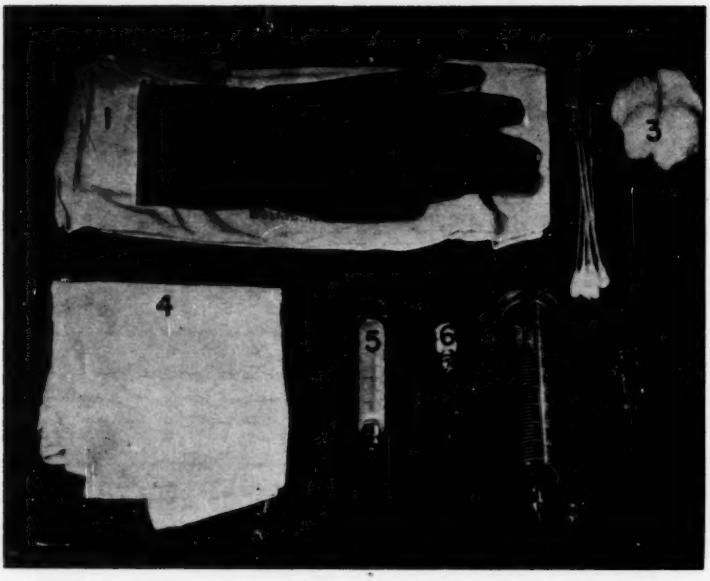

FIG. 1A. Sterile equipment used in performing aspiration. (1) Sterile gloves; (2 and 3) materials for preparation of skin; (4) spinal drape; (5 and 6) syringe and needle for novocain infiltration; (7) 30 cc. syringe used in performing aspiration; (8) Klima Schleicher needle.

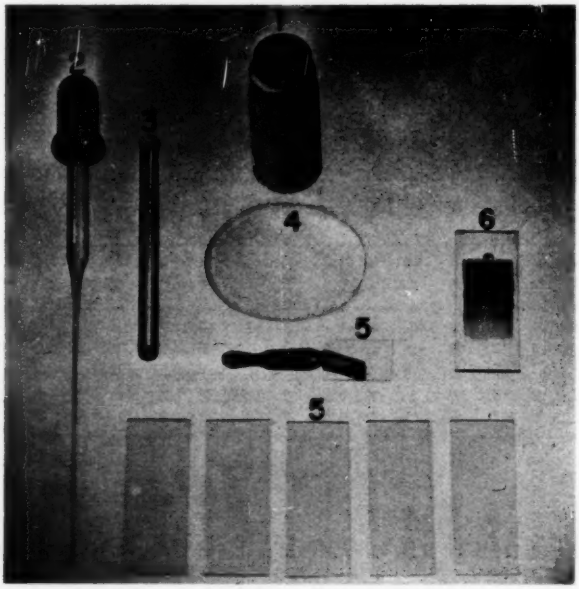

FIG. 1B. Equipment used in obtaining volumetric data and in preparation of microscopic slides. (1) Paraffin-lined vial containing heparinized marrow; (2) pipette for use in filling hematocrit tube and aspirating ME after centrifugation; (3) hematocrit tube; (4) paraffin-lined watch glass onto which ME and equal volumes of plasma and red cells are delivered and mixed; (5) slides and finished smear.

Most bone marrow aspirations performed on the surgical cases were done immediately after operation; occasionally aspiration was done pre-operatively. The anesthetics used at operation were for the most part intravenous pentothal or open drop ether. Surgery averaged approximately 40 min. in duration and ranged from 5 min. to 1½ hr.

Equipment and Supplies: The items of equipment employed in the technic are illustrated in figures 1A and B, and the procedure is explained in the legend. In order to preserve maximum cytologic detail, powdered dog liver heparin* is used as anticoagulant rather than oxalates. The needle used in all aspirations, including the newborn infants, was the standard-size Schleicher type.† The adaptor shown in the illustration is a vital part of the apparatus; it insures an airtight connection between syringe and bone marrow needle. Syringes of 20 to 30 cc. capacity were used for the aspiration. They must be absolutely airtight.

Site of Aspiration: Tibial aspirations were performed on 17 newborn infants and 23 infants and

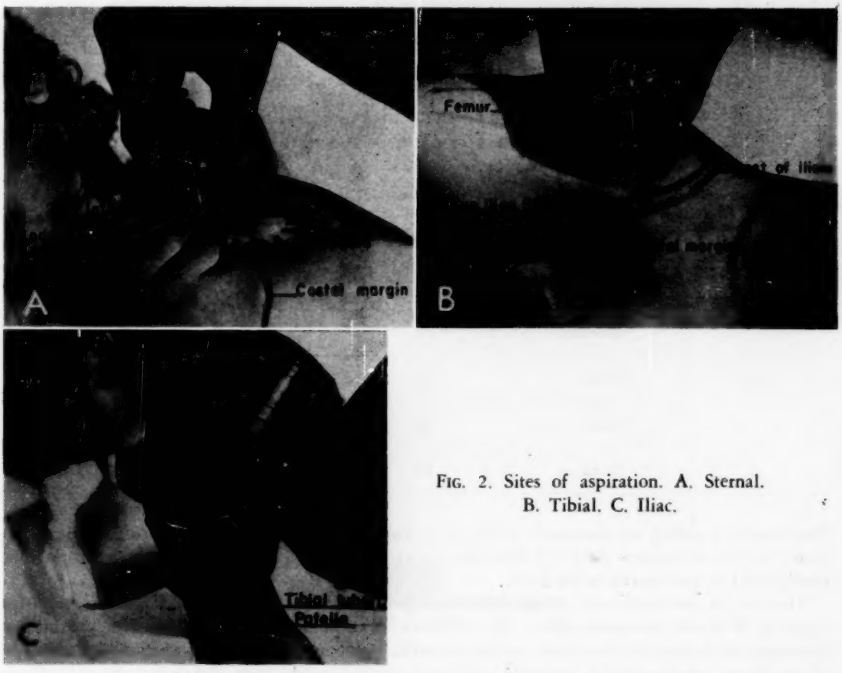

FIG. 2. Sites of aspiration. A. Sternal. B. Tibial. C. Iliac.

children, 2 wk. to 5 yr. of age. Sternal aspirations were performed on 10 children from 5 yr. through 11 yr. of age. On all the 22 children over 17 mo. of age, simultaneous iliac aspirations were performed.

The 3 sites of aspiration used in this study are illustrated in figures 2A, B and C. The tibial site was located above one half the distance between the tibial tubercle and the medial margin of the tibia, at a level slightly below the tibial tubercle. Sternal aspiration was performed in the midline at a level between the second and third ribs. The iliac site was approximately 3.8 cm. posterior to the anterior superior iliac spine and 2.5 cm. below the superior margin of the iliac crest. In all instances the needle was directed perpendicularly to the plane of the bone at the site of puncture. In the case of the iliac puncture, this plane could best be estimated while infiltrating the periosteum (or by probing with the aspiration needle). This was accomplished by determining the depth of the ilium beneath the skin in several directions.

Sedation: Most of the patients did not require preliminary sedation inasmuch as the procedures were performed immediately after operation. However, under ordinary circumstances it has been found

advisable to give some sedation routinely to all infants and children over 12 mo. of age. Morphine sulfate in the dose of 2 mg./10 lb. body weight ½ hr. prior to undertaking the procedure has proved superior to other sedatives and analgesics.

Procedure

With the operator wearing sterile gloves, the skin at the site of the aspiration is prepared with 1/1000 tincture of merthiolate. If the patient shows signs of arousing from the general anesthesia, the skin, subcutaneous tissue, fascia and periosteum are thoroughly infiltrated with about 2 to 5 cc. of 1% novocaine. The needle guard is adjusted to the approximate depth of the bone beneath the skin, and the soft tissues are penetrated with the needle. In puncturing the bone, firm pressure is applied and a simultaneous rotating and wedging motion used until the needle feels firmly impinged in the bone. Then, with continued firm pressure on the guard and simultaneous rotating of the needle through approximately 180-360° (clockwise and counterclockwise), a vague "give" is usually felt and the patient often manifests slight pain; the puncture is completed.

The stylet is withdrawn, the syringe and adaptor tightly affixed, and the aspiration performed.

TABLE 2

Average and Range of M.E. and Fat in mm. from Tibial, Iliac Crest and Sternal Aspirations

	Site	No. of Cases	Average	Range
Me	Tibia	39	4.7	1 to 11.0
	Ilium	22	4.1	1 to 7.0
	Sternum	10	3.8	2 to 7.0
Fat	Tibia	39	2.3	0 to 6.0
	Ilium	22	1.0	Trace to 2.0
	Sternum	10	1.0	Trace to 3.0

The plunger is pulled out vigorously to the 25 cc. mark and held there for 5 to 20 sec., during which time 2 to 5 cc. of marrow fluid will flow into the syringe except from the tibiae of newborn infants, where only 1 cc. can usually be aspirated.

The fluid is discharged into the paraffin-lined tube containing the heparin and agitated gently for 1 min. A Wintrobe hematocrit tube is then filled with 1 cc. of the fluid, centrifuged at approximately 3000 rpm for 6 min., and the height of the several layers noted. The fat layer, which floats to the top of the plasma, on occasions is composed of 2 parts, an upper white layer of free fat cells and a lower red layer of bone marrow tissue. Between the plasma column and the column of packed red cells at the bottom of the tube is a reddish-white layer consisting of myeloid and erythroid cells derived from the bone marrow (the M.E. layer). The height of the several layers is recorded as the volumetric pattern of the marrow.

Further processing of the contents of the hematocrit tube and of the remaining bone marrow fluid for microscopic study will be described in subsequent reports.

Results

Table 1 lists the results obtained from each aspiration.

For the most part, surprisingly little technical difficulty was experienced. In all aspirations, excepting those performed on newborn infants, two or more cubic centimeters of fluid were obtained easily on the first puncture. The marrow flowed into the syringe in

from 3 to 10 seconds. No complications of any significance occurred. Of the 17 aspirations attempted on the newborn infants, only in one instance (Case 8) was insufficient fluid obtained. In two more instances (Cases 5 and 9), less than 1 cc. was obtained. In the remaining 14 aspirations, 1 to 2 cc. of fluid were obtained at a slow rate (20 seconds). In one instance 3 cc. were obtained easily. In general, 3 to 6 day old infants offered less difficulty in this regard than did those of 1 to 2 days.

With the use of the described technic, the tibia proved a satisfactory site for the study

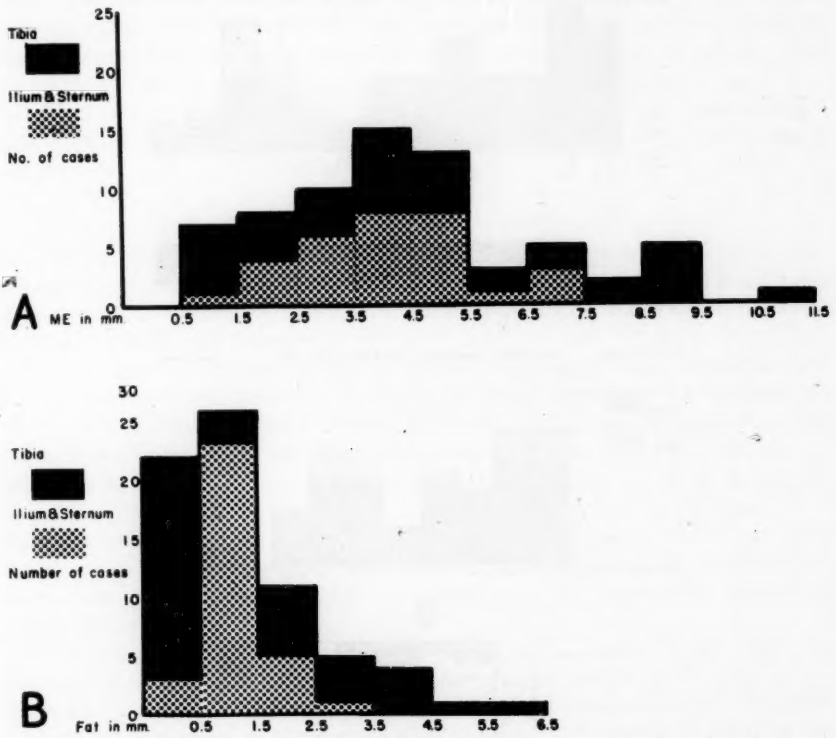

CHART 1. Distribution of ME and fat values comparing combined sternal and iliac values to tibial value. A. ME. B. Fat.

of bone marrow in infants under 18 months of age; subsequent to 18 months of age, the iliac crest proved uniformly satisfactory. Sternal aspirations were not attempted on individuals below the age of 5 years because previous experience had indicated that the procedure could be very trying for both patient and operator.

Charts 1 A and B show the distribution of "myeloid-erythroid" and fat values, respectively, from the combined sternal and iliac aspirations in contrast to the tibial aspirations. The myeloid-erythroid values range from 0.5 to 11.5 mm.; in no instance was there an absence of the myeloid-erythroid layer. The fat values range from 0 to 6 mm.

Charts 2 A, B and C show the influence of age on the myeloid-erythroid and fat layers obtained from the tibial aspirations, the iliac aspirations and the sternal aspirations,

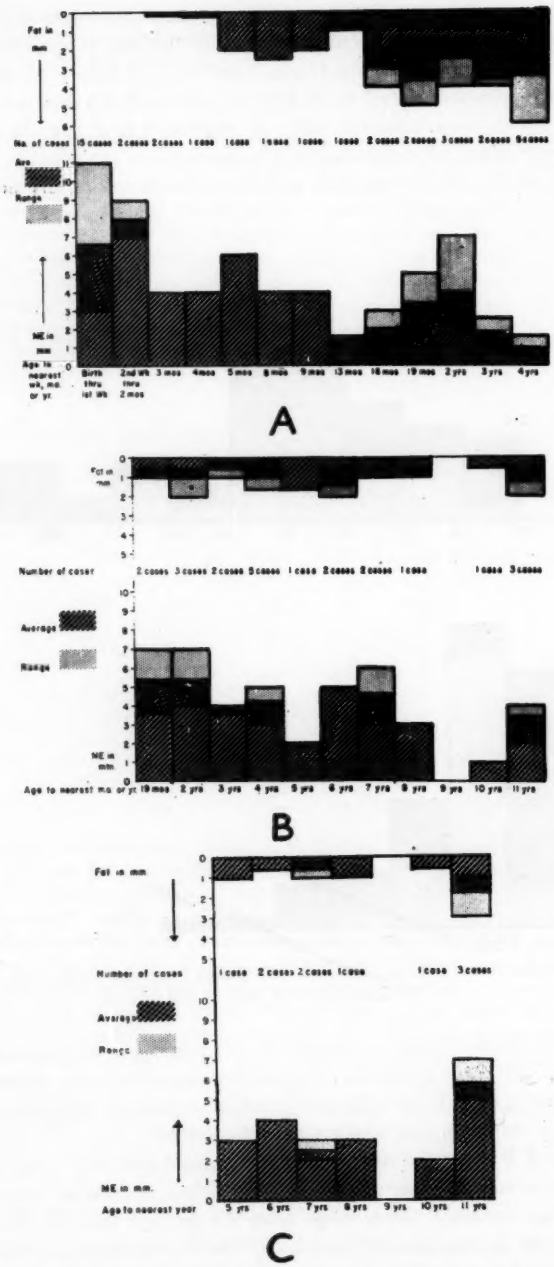

CHART 2. Influence of age on ME and fat values. A. Tibial aspirations. B. Iliac aspirations. C. Sternal aspirations.

respectively. In the case of the tibial aspirations, it should be noted that with increasing age there was a general tendency for the fat to increase and the myeloid-erythroid to decrease. By 3 years of age the myeloid-erythroid was reduced to a low value and the fat became quite abundant. The five aspirations performed in the 4 year old range yielded uniformly very low myeloid-erythroid values; and even as early as 16 months of age, tibial aspirations yielded low myeloid-erythroid values (1 mm.) and quite high values for fat (4 mm.). In contrast to the tibia, the iliac and sternal sites did not show the distinct decrease in myeloid-erythroid and increase in fat after 3 years of age. Aspirations from these latter two sites resulted in values that tended to parallel each other. Although iliac aspirations were not performed on the infants under 18 months of age, there is a suggestive increase in myeloid-erythroid in the individuals below 3 years of age.

Table 2 lists the average and the range for fat and myeloid-erythroid values obtained from the three sites studied. An average value for the tibia serves little purpose inasmuch as there is such great variation with age.

Discussion

It will be noted that in some instances clinical diagnoses are such that one might conceivably expect some response of the hemopoietic tissue to the disease process listed. Likewise, peripheral blood values cannot be considered to be strictly within the normal limits as listed by Blackfan and Diamond.[17]

Case 22, listed as intussusception, had minimal symptoms of bowel obstruction and was explored one week previously. At surgery no changes in the bowel were noted and the convalescence was uneventful. Several of the cases studied had total white blood cell counts over 10.0 thousand/cmm. For the most part these occurred in infants. In this age group almost all the total white counts were elevated; the counts were taken immediately on entry of the patients to the hospital, at which time the infants are frequently in a state of considerable excitement. In the older age groups the white counts over 10.0 thousand/cmm. are not associated with an increase in the percentage of polymorphonuclears; hence, they are not considered manifestations of an inflammatory process, but rather normal variations.

Technical Considerations: The adaptability of the technic described in this report for the study of infants' and children's bone marrow was questionable at the outset of the study. One can obtain so little bone marrow fluid from the sternum of infants (less than 1 cc.[1-3]) that different sites of aspiration had to be used. With the use of the tibial site below 18 months of age, sufficient fluid was obtained for study with the preceding technic in all but three instances.

Numerous reports of bone marrow studies mention that the aspiration of fluid in excess of 0.2 cc., or at most 1 cc., result in dilution of the original specimen with sinusoidal blood. To the contrary, Osgood and Seaman[13] state that values obtained from aspirations between 1 to 10 cc. in quantity yield more uniform results than do those from studies of the smaller quantities. They did not note "dilution effect" when large quantities were aspirated. Vaughan and Brockmyere[15] likewise recommend the use of 3 cc. samples. In the present study the maximum amounts of fluid aspirated (5 cc.) did not yield uniformly low myeloid-erythroid values but showed a spread of values almost comparable to those found when 2 cc. were aspirated.

Nucleated cell counts per cubic centimeter of fluid were not performed in this study.

The myeloid-erythroid value, of course, furnished essentially the same information. This value is obtained more easily and rapidly and is not subject to as much variation; that is, the variables in a hematocrit determination are smaller and fewer than those in a hemocytometer chamber count. However, it should be mentioned that bone marrow samples having late normoblastic hyperplasia did yield a myeloid-erythroid layer almost identical in color to the red cell layer. Under these circumstances one experiences considerable difficulty in reading the line of junction between the myeloid-erythroid and the red cell layers. The hematocrit determination also has the very distinct advantage over the hemocytometer chamber count technic that, in addition to the hemopoietic cellularity (M.E. or nucleated cell count), the fat value furnishes definite information regarding the percentage distribution of fat and myeloid tissue in the marrow. In general the fat values found in this study of tibial marrow bear a reciprocal relation to the myeloid-erythroid value. Thus with an hematocrit determination an additional value is obtained that may be useful in forming an opinion of the histologic nature of the bone marrow. Studies now in progress in pathologic conditions show a better correlation between the amount of volumetric fat and the fat tissue seen in histologic material than between the quantity of myeloid-erythroid layer and the histologic cellularity.

Influence of Age on Values Obtained from Different Sites of Aspiration

Tibia: Frequent reference is found to the study by Custer and Ahlfeldt,[18] who, in 1932, reported gross observations on sections of tibias and other bones obtained at postmortem from "unselected" infants, children and adults; the cause of death is not mentioned. They demonstrated that yellow fatty marrow did not appear in the tibia until 7 years of age. Such observations can hardly be considered normal unless they were obtained from healthy infants and children who died sudden traumatic deaths.

Piney,[19] in 1922, reported on the macroscopic observation of bone marrow from 91 subjects, 3 days to 83 years old, in previous good health, who had died suddenly. He observed that the cut surfaces of the long bones became gray as age 7 years is approached and that by 12 to 14 years a patch of gross fat became visible in the middle of the shaft. He realized ". . . that red tissue in the medullary cavity is not necessarily true cellular red marrow. Microscopic examination is essential for determination of the limits in normal living infants and children." The volumetric data found in this study suggests that the tibial marrow becomes relatively fatty and acellular as early as 16 months of age and practically uniformly so by 3 years of age. Study of histologic material and differential cell counts, to be reported subsequently, lend confirmation to this impression.

Iliac Crest: In general, iliac crest aspiration by others has not been performed as routinely as sternal aspirations. Several reports, however, have been made on the use of this site for bone marrow aspiration.[8, 20, 21] Originally Van den Bergh[20] and recently Rubinstein[21] and Rheingold et al.[8] have pointed out several advantages of the iliac site over the sternal site; the experience gained in the present study amply confirms their experiences. The quantitative data from the two sites appear to be comparable, but the iliac crest aspirations are more easily performed from the standpoint of operator and patient. The iliac crest values, in contrast to those obtained from the tibia, do not show a marked change with increasing age.

Sternum: The obvious hazards of sternal aspiration are substantiated by the reports of Rubinstein,[21] Schorer[22] and Meyer.[23] In the present study sternal aspirations were

not performed on children under 5 years of age; the volumetric data obtained from the 10 punctures performed on children from 5 years of age through 11 years showed essentially the same values as Schleicher[10] reported in 14 males 18 to 40 years of age. For fat, his chart showed a trace to 3 mm. with an average of 1 mm. The same values were found in the present series. He reports the myeloid-erythroid as varying from 4 to 6 mm. with an average of 5 mm. The present results show a wider variation, that is, from 2 to 7 mm., with an average approximately 4 mm. With the methods employed in this study of children 5 to 11 years old, age was not shown to influence the values from sternal aspirations.

Conclusion

The volumetric technic described is technically practical for the study of bone marrow in infants and children.

Tibial aspirations show in general a progressive increase in fat and decrease in myeloid-erythroid layer with advancing age; after approximately 2½ years of age, many of the specimens reflect a relatively fatty and hypoplastic marrow.

Iliac aspirations from individuals 18 months through 11 years of age yield volumetric values comparable to the values obtained from sternal aspirations.

Sternal aspirations from 5 years through 11 years yield values similar to those reported in the older age ranges.

With use of the technics described, the iliac site is technically superior to the sternum for the study of bone marrow of children from 18 months through 11 years; for those individuals less than 18 months of age, the tibia offers a practical site.

Acknowledgment

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SPANISH ABSTRACT

Caracteristicas Volumetricas y Microscopicas de la Medula Osea en Infantes y Niños

I. Caracteristicas Volumetricas

El autor estima que el metodo volumetrico descrito es técnicamente práctico en el estudio de la medula ósea de infantes y niños.

La aspiración de medula ósea de la tibia demuestra en general un aumento progresivo en grasa con disminución de la capa (layer) mielo-eritroide del hematrocrito con el avance en años; y aproximademente después de los 2½ años de edad muchas de las aspiraciones demuestran una medula ósea relativamente grasosa y hipoplástica.

Las aspiraciones del ileum entre los 18 meses y 11 años de edad revelaron valores volumetricos comparables a los obtenidos con aspiraciones esternales.

Las aspiraciones del esternón entre los 5 y 11 años de edad revelaron valores similares a los reportados en los adultos.

El autor sugiere que mediante el uso de las técnicas descritas, el hueso iliaco es técnicamente superior al esternón para el estudio de la medula ósea en niños entre los 18 meses y 11 años de edad. En niños menores de 18 meses la tibia ofrece el sitio más práctico.

4614 Sunset Boulevard

Pediatrics

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AMERICAN ACADEMY OF PEDIATRICS, INC. PROCEEDINGS AND REPORTS

THE PRESIDENT'S PAGE

ONE of the first acts of this administration was to write a letter to those foundations and commercial firms which had donated so generously to the work of the ICH Committee. In that letter it was pointed out that previously the Survey Committee had been assigned a task and when it was completed the committee was dissolved. They turned their data over to another group for them to discover the significance of the facts that had been gathered. This was the Committee for the Improvement of Child Health. They, in turn, had completed their task and had been dissolved. Their work had been tremendous and their influence great. They had brought the Academy a leadership in the field of child health which it had never before realized.

The letter further stated that the task of the Academy now was to carry on the work of the ICH Committee and to build on the foundation that had been so well laid. It was our purpose to integrate its work among the several committees and, thereby, not only have all the membership share the responsibility, but increase the usefulness of our organization by the participation of more members.

The response to the letter was almost unanimous and showed a deep interest in our work. A number stated that their budgets were allocated for the year, but they would be glad to consider us if we should later send in a specific request. Others sent us money. All were grateful for the information and expressed their belief that the Academy was a power for better child health.

This year our commission, then, is to see to it that we not only carry on, but expand the work that the ICH Committee began. I can report to you even now some of our present activities which have been initiated.

1. The second volume of the Interpretive Report, which will include a chapter on community planning, will be published as a supplement to PEDIATRICS.
2. The full report of the 1950 Educational Regional Conferences will be published as a supplement to PEDIATRICS.
3. A summary of the child health work in the States, which was a result of or stimulated by the survey, will be published.

4. The regional meetings of pediatric educators are being continued.
5. Summaries will be made and published of regional pediatric education throughout the United States and Canada.
6. A panel and exhibit on pediatric education will be a part of the annual meeting.

Beyond the work of the ICH Committee we have already accomplished the following:

1. Established at the central office an Information Bureau to enable the chiefs of pediatric departments to get in contact with pediatric residents for their staffs and to enable prospective house officers to find openings in hospitals.
2. Laid the plan to publish a pamphlet to the membership on the hospital care of the child, which will give concisely modern ideas on all phases of that subject from correct hospital construction to the psychological care of the bed-ridden.
3. Arranged for a pamphlet to be published to all physicians of the United States calling attention to the diagnostic and therapeutic facilities available for neoplastic disease in children.

This is not a complete summary of what we have done. It is not intended for that. It is to illustrate the fact that we are continuing the work of the ICH Committee, we are building on their experience, and we are expanding our activities. These activities are now in the framework of the whole Academy, and our goal of greater child health leadership for our organization is being realized.

PAUL W. BEAVEN, *President*

Pediatrics

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SPECIAL SECTIONS

THE PEDIATRICIAN AND THE PUBLIC

AN OPEN FORUM

PAUL A. HARPER, M.D., *Editor*

Statements appearing in this column do not necessarily reflect the opinion of the editor nor are they to be interpreted as the official opinion of the Academy.

COMMUNICATIONS FROM DR. MARTHA W. ELIOT AND DR. LOUIS VERHOESTRAETE

DEAR DR. HARPER:

Needless to say, I am most happy to forward to you a communication from Dr. Louis Verhoestraete on a subject which has long been of deep personal concern to me. From his letter it will be apparent that the World Health Organization attaches very great importance to its work in the field of maternal and child health, and Dr. Verhoestraete has explained in some detail the methods and philosophy behind the international activities of WHO in its work for children.

Dr. Verhoestraete is a Belgian paediatrician with considerable international experience. He received his early medical and paediatric training in Belgium, The Netherlands and The United Kingdom. He carried on a private paediatric practice in Antwerp and was Medical Adviser to the Belgian National Committee on Children's Health from 1942 to 1947. In this latter year he was sent on a mission by the Ministry of Public Health in Belgium to study child health administration and modern paediatrics in the USA. At the end of eight months, in 1947, he joined the WHO staff as paediatric consultant to UNICEF in Paris, and since 1950 he has been Chief of the Maternal and Child Health Section at WHO headquarters in Geneva, Switzerland.

MARTHA M. ELIOT, M.D.
Geneva, Switzerland

DEAR DR. HARPER:

In accepting your invitation to prepare a communication for PEDIATRICS describing the Maternal and Child Health activities of the World Health Organization, I am acutely aware of the responsibility which I have undertaken, and at the same time I am very

happy to have this unusual opportunity of "talking over"—through your open forum—the ways in which the World Health Organization is attempting to improve the health of mothers and children throughout the world.

In the first place, the scope of the problem faced by any "world" health organization is tremendous. Take, for example, child health. About one third of the world's population consists of children, but less than one tenth of the world's population lives in areas where the known infant mortality rate is as low as 50 deaths per 1000 live births. Only about one quarter of the countries in the world have an infant mortality rate below 50. In the United States, the infant mortality figures for 1949 lie roughly between 25 in a few areas to somewhat over 40 in others. In the world at large the infant mortality rates by countries, range roughly between 25 and an estimated 400, or even higher. These widely divergent rates reflect the great contrast between the economically more advanced and the economically underdeveloped countries. The incidence of the three chief causes of infant death in several parts of the world in 1946—the year the World Health Organization was formed—is listed in the following table:

	Infant Mortality Rates/1,000 Live Births 3 principle causes			
Countries	Totai	Gastroenteritis	Congenital Malformations, Prematurity and Debility	Bronchitis, Pneumonia and Influenza
Egypt	187.1	104.2	59.2	16.5
Chile	159.5	25.1	52.7	48.0
Portugal	119.4	46.6	34.5	12.8
Italy	86.9	20.1	32.7	20.6
Netherlands	38.7	3.3	19.4	4.5
USA	33.8	1.7	23.3	3.8
Australia	29.0	1.0	21.3	3.2
Sweden	26.5	.7	15.3	3.8

Unsanitary conditions prevail for three fourths of the world's people, who dispose of their wastes dangerously; are plagued by rodents, insects and parasites; drink unsafe water. Poor lands, lack of cattle and lack of knowledge lead to chronic malnutrition. In some regions the whole population may be affected by such epidemics as cholera, dysentery or typhoid fever. In some tropical and subtropical countries malaria alone causes an estimated 3 million deaths each year, taking heavy toll of infants and young children. Of the estimated 2 million active cases of yaws among the inhabitants of tropical areas, 75% are children under 15 years of age. Eighty-five per cent of the children attending schools in some areas are afflicted with trachoma. Tuberculosis and venereal infections are widespread.

This is a terrible picture of the problems of life facing more than half the children of the world. But death and sickness are not the only problems. Not only must the child survive, but there is a need to foster in him the normal processes of growth and development so that he may be physically well, emotionally mature, mentally alert, and be able to live as an adult in a satisfactory relationship with his family, the community and the world outside. This is true for all children, but has been increasingly emphasized

in the more economically advanced countries where hazards to healthy personality development in the child are increased due to greater emotional stress and strain put on family life by problems of high industrialization. The emphasis placed on these questions at the recent Mid-Century White House Conference on Children and Youth, which I was fortunate to attend as a WHO observer, illustrate this concern.

Such considerations are implied in the concept of "health" which this world organization holds. It was not many decades ago that "international health" consisted of the erection of quarantine bulwarks by individual nations against the invasion of pestilential diseases from abroad. Then, largely through stimulation given by the League of Nations, quarantine and other international health regulations were increasingly developed. Today, however, the concept of international health is much broader. To emphasize this new concept, let me review the historical beginnings of WHO.

The United Nations Conference on International Organizations held in San Francisco in 1945 recognized the vital importance of health as a factor in the promotion of conditions of stability and well-being for the nations of the world, and health was, therefore, included among the fields of co-operative endeavour with which the United Nations should concern itself. As a result of decisions made at the San Francisco Conference, an International Health Conference met in New York City in June 1946. At the closing ceremonies of this Conference, the Constitution of the World Health Organization was signed on behalf of 61 nations, and an Interim Commission was created to lay the groundwork for the establishment of the permanent World Health Organization.

The Constitution which was thus signed in New York was a truly revolutionary document. Its significance lies in its definition of health now familiar to us all—"a state of complete physical, mental and social well-being and not merely the absence of disease or infirmity." The Constitution also affirms that health, thus defined, is "one of the fundamental rights of every human being," and it recognizes that the "health of all peoples is fundamental to the attainment of peace and security and is dependent upon the fullest co-operation of individuals and states." With equal emphasis the Constitution also maintains—"Healthy development of the child is of basic importance; the ability to live harmoniously in a changing total environment is essential to such development." These broad health concepts are the foundation on which the World Health Organization is built.

A far-reaching "organization" of effort was therefore needed to make these world health principles become a practical reality. The Constitution was finally formally ratified by the necessary 26 States Members of the United Nations in April 1948, and in September of that year the World Health Organization became a permanent autonomous body. It has now a total membership of almost 75 countries in all parts of the world, and is one of the largest among the specialized agencies of the United Nations. Representatives from these Member States meet every year at the World Health Assembly to discuss programme plans and budget. The Fourth World Health Assembly will convene in Geneva, Switzerland, in May 1951, and will put forward proposals for the work of the Organization through 1952.

During its first four years—two as an Interim Commission and two as a Permanent Organization—WHO has grown and developed, and has taken a positive approach in assisting countries to achieve better health for their people. The Regular Budget of WHO, representing contributions from Member States according to a scale established by the

Assembly, is very small in proportion to the task—$5 million in 1949, $6.3 million in 1950, $6.15 million in 1951. In 1951 the Regular Budget is being supplemented by funds made available to WHO from the United Nations programme for Technical Assistance for economic development. Like the other specialized agencies of the United Nations, it was never intended that WHO should be primarily a "supply" organization. Practically all WHO's funds go to the task of rendering technical and advisory services to countries, toward the establishment of a wide variety of technical standards, and the collection and dissemination of health information and morbidity and mortality statistics.

First, WHO has assumed certain technical responsibilities which are so truly international in scope that they can be administered and carried out most effectively from a central Headquarters in Geneva, Switzerland. Some of these responsibilities are the so-called "traditional" services of international health which were taken over by WHO from the League of Nations—e.g., epidemiological intelligence, and biological standardization —and from the Office International d'Hygiène Publique, e.g., revision of international sanitary conventions. Services such as these, benefiting nonmember countries of WHO as well as member countries, are supplemented by other activities along similar lines—e.g., Unification of Pharmacopoeias, revision of the International Lists of Diseases and Causes of Death; adoption of international regulations on health statistics, establishment of research projects on parasitic, virus, and bacterial diseases; development of international standards for such procedures as laboratory methods; preparation of international health publications; and planning of world-wide and pilot studies on various phases of public health in co-operation with other agencies.

Second, WHO is supplying advisory aid to national governments in order to encourage and help countries to develop and strengthen their own health services. This strengthening of health administrations of member governments is of basic and fundamental importance to the long-term improvement of over-all health conditions in any given country, and it is also essential for the success of the above-mentioned international health activities. The type of health projects which may be advisable for a country to initiate and develop will depend on the particular health problems, the economic situation and the availability of technical personnel in the country—but the long-term objective is a balanced programme with emphasis on prevention of disease and promotion of health.

In order to bring its advisory services closer to the individual countries the Constitution of WHO lays down the broad outlines of a regionalization plan. WHO now has three fully established regional offices, one temporary regional office, and two special offices serving two regions not yet completely organized. The oldest and most fully developed of these is the Regional Office for the Americas, located in Washington, D.C. In 1947, steps were taken to utilize the Pan American Sanitary Bureau, established in 1902, as the future WHO Regional Office. This integration is going forward rapidly and the WHO PASB Regional Office in Washington is an active, well-staffed centre where health problems of all the Americas are studied with insight and understanding of the needs and wants of the individual countries. A Regional Office for South East Asia was established in New Delhi, India, in 1948. A Special Office for Europe was set up in Geneva in 1949, and a Regional Office for the Eastern Mediterranean area was organized in Alexandria, Egypt, in the same year. In 1950, the Temporary Regional Office for the Western Pacific was established in Hongkong, and an Office for Africa has begun its work from temporary headquarters, in Geneva.

Although administratively linked to WHO Headquarters in Geneva, each regional office has great individuality, developing its assistance along lines dictated by the needs of its member countries. However, the Department of Advisory Services, operating from WHO Headquarters in Geneva, has the specific task of supporting and providing technical guidance for the regional programmes, of co-ordinating the work on a world-wide basis, and of assessing the results. This Department consists of three Divisions, viz., the Division of Organization of Public Health Services, the Division of the Control of Communicable Diseases, and the Division of Education and Training Services. In 1948 a special Section on Maternal and Child Health was established and has been included in the Division of Organization of Public Health Services along with other Sections having related interests, e.g., public health administration, nursing, health education, social and occupational health, nutrition, mental health and sanitary engineering.

To meet the health needs of almost 75 different countries with wide divergence of cultural and economic backgrounds is not in the long run a problem than can be solved by one international health agency working alone. It involves many questions of social welfare and education and is certainly dependent on the economic conditions within the countries. Thus a co-ordinated approach by the United Nations Division of Social Activities and the specialized agencies concerned is necessary—for example, United Nations Educational Scientific and Cultural Organization (UNESCO), Food and Agriculture Organization (FAO), the International Labour Office (ILO). These agencies, like WHO, provide technical advice and advisory services at the request of governments. Another agency, the United Nations International Children's Emergency Fund (UNICEF) has a different and yet particularly important co-operative role in connexion with the maternal and child health work of WHO. UNICEF was established by the United Nations on a quite different basis from the permanent specialized agencies mentioned above, in that it is a "Fund" and is basically concerned with providing supplies and equipment. During its first years of activity, the major part of its funds were spent in relief programmes for children of war-devastated countries. Now an increasingly larger portion of UNICEF funds are being spent for the purchase of supplies and equipment for health programmes developed with WHO, within the policies laid down by a Joint Committee on Health Policy of the two organizations. These joint health programmes on behalf of children include, in addition to specific maternal and child health programmes, tuberculosis control, BCG vaccination, venereal disease control programmes including yaws, and malaria and insect control programmes. Such joint WHO/UNICEF programmes can go far beyond the local "demonstration" phase, because UNICEF funds for supplies and equipment make possible large-scale projects even on a country-wide basis.

It is also necessary that WHO and certain nongovernmental voluntary associations in technical or welfare fields should find ways of co-operating to produce the best long-lasting results. Certain recognized international voluntary organizations, including the newly organized International Pediatric Association, have been admitted into a formal working relationship with WHO. Co-operation and co-ordination of this sort is of paramount importance and WHO is fostering and encouraging this approach.

This is the historical and organizational framework of the World Health Organization. Into this framework have been built ideas and programmes of action, based on experience and technical knowledge, with which to attack the vast and divergent problems of health never before attempted to the same extent on a world-wide scale. The health

needs of the child have been considered at every stage of the planning, since one of the chief constitutional functions of WHO is "to promote maternal and child health and welfare."

Most of the programmes of WHO have a bearing on maternal and child health. In order to deal with the over-all problems and diseases in countries where vast health problems exist, building up the general public health services is most essential. In these underdeveloped areas a programme of special services for mothers and children cannot, in itself, meet their needs.

One of the most important steps taken by WHO to strengthen general public health services is a vigorous educational programme. Knowledge and technical skill required to save lives and promote health are available in the world to-day, but what is most urgently needed is more trained people to use this knowledge. In most countries of the world a shortage of medical and para-medical help is reported. A simple comparative observation between two countries may illustrate the extent of this shortage in some areas.

	United Kingdom	Ratio/ population	India
Doctors	1-1,000	:	1-6,300
Nurses	1-300	:	1-43,000
Health Visitors	1-4,770	:	1-400,000
Midwives	1-618	:	1-60,000
Pharmacists	1-3,000	:	1-4,000,000
Dentists	1-2,700	:	1-300,000

WHO's programme of education and training of medical and related health personnel is sponsored by the Division of Education and Training Services. It is varied, and adaptable to the level of scientific and social knowledge in a country. Group training opportunities are stimulated in countries; institutes of training are set up; reforms in basic professional and technical education are encouraged; medical libraries are provided with literature and teaching equipment; exchange of scientific information is furthered; ways of training and utilizing auxiliary workers are studied and applied; individual fellowships are granted to doctors, health officers and nurses. In the field of maternal and child health alone, approximately 80 fellowships have been awarded in the past four years to recipients from about 20 different nations. A general policy in regard to these fellowships is that they shall be given to those candidates who are, or may become, responsible for public health administrative activities within their own country; or for those who are, or may be, in charge of teaching programmes in medical centres. Many of these fellowships are given in connexion with definite WHO projects being undertaken in member countries.

Another method of strengthening the general public health services of a country is through the public health advisers, attached to WHO Regional Offices. These advisers study the health needs of countries in their regions and plan with them what measures should be taken. However, any activities with which WHO is to assist a country must be desired and requested by the Government. They must be activities which the countries themselves will initiate and which they will carry on after WHO has withdrawn. This is of extreme importance since the necessity of adapting techniques and methods to local conditions and cultures is obvious, and since superstitions, customs, religious taboos and pride may seriously impede the introduction of new health ideas unless wisely handled.

The national or local activities to which WHO gives assistance are, as a rule, those for which reasonably satisfactory techniques are well established, and from which large numbers of the population will derive real benefit. Though improvement of the national health services is the broad objective, it may be necessary to begin with projects in a specialized field which will start immediate, extensive and easily recognized improvements, such as malaria control by DDT spraying, sanitation, nutrition and food production, yaws eradication, tuberculosis control, or maternal and child health. Although these general and specific activities of public health work are not necessarily directed exclusively towards improving the health of children, nevertheless, they are quite obviously fundamental to the protection of child health. Where such programmes for the improvement of the general level of health have been carried out in countries by WHO, or jointly with UNICEF, the child population has benefited enormously.

But along with the organization of general public health services, a more direct approach to the problems of the mother and child must be undertaken, where appropriate. To help in this more direct approach, maternal and child health advisers have been attached to the Regional Offices of the Americas, Eastern Mediterranean, Europe, South East Asia, and the Western Pacific. These maternal and child health advisers like the advisers in public health administration, nursing, and others, are available to the Regional Director to help in the planning of the health activities in the area, including the specific maternal and child health regional programmes. These maternal and child health advisers are also available to assist the governments in their areas. During 1949 and 1950, they have surveyed, visited or made studies in most of the countries of their regions, and reports on health conditions of mothers and children from all parts of the world have reached headquarters in Geneva. With this background and knowledge, the Maternal and Child Health Advisers have been prepared to help national administrations make concrete plans towards the development of their services for mothers and children.

Many countries are not ready for the implementation of maternal and child health programmes on a large scale. Generally speaking, the programmes developed with the governments in the regions of South East Asia, Western Pacific, Eastern Mediterranean, and to a lesser extent in South America, are in the nature of demonstration and training projects. Such projects are set up with the purpose of helping countries to establish and strengthen their own maternal and child health services as a part of the development of their over-all public health structure. These urban or rural projects serve to demonstrate sound maternal and child health practices. They include the establishment within a prescribed area of pre- and post-natal clinics, child health centres, and school health services, together with improved clinical facilities in the community, such as more adequate pediatric and obstetrical clinics. They are to be used as training fields for schools of public health in the area and for teaching preventive pediatrics to under-graduate and post-graduate medical students. They are to serve in the training of nurses, public health nurses, midwives, nurse midwives; and for the special type of training for young women who will provide auxiliary aid to nursing services. Obviously the training of nursing personnel in rural areas is of utmost importance.

In this connexion it is significant that the first maternal and child health demonstration and training project which received WHO assistance is located in Najafharh, a rural area near New Delhi, India. This project has been particularly concerned with building up a training base for different kinds of nursing and auxiliary personnel.

At the request of the government, teams of international personnel are attached to these demonstration projects in an advisory capacity. These teams are made up generally of one maternal and child health medical officer, and one or more public health nurse tutors and midwife tutors. It is WHO's particular concern that the countries themselves take the initiative in these projects by employing national and local personnel who, with the co-operation and guidance of the international team, will gradually assume complete responsibility for the project. This is, in WHO's opinion, the surest safeguard that the work will continue. In order to assist in the development and training of national leaders in maternal and child health, it has been found useful in a number of instances to offer WHO fellowships to selected professional workers connected with the programme.

Projects of this general type have been started in New Delhi, India; in Manila, Philippine Islands; in Lahore, Pakistan; in Kabul, Afghanistan; and in Rangoon, Burma. Still others will be initiated during 1951 in Indonesia, Thailand, Ceylon, Formosa, Paraguay, Peru, Colombia, Chile, Ecuador, Pakistan, Iran and Syria.

A few projects are exclusively concerned with teaching maternal and child health practices to nurses and auxiliary health personnel. In Malaya, Ceylon, Sarawak, Brunei and North Borneo internationally recruited nurses experienced in maternal and child health are now teaching in centres and hospitals, and helping to establish adequate local facilities for training nurses, midwives and other auxiliary health workers. WHO has also, in some cases, taken advantage of the existence of a field project of a different nature to stimulate an awareness of maternal and child health practices to new areas. This has been achieved, for instance, by assigning public health nurses with maternal and child health experience to WHO venereal disease and malaria teams.

Convinced of the enormous importance of the training of well qualified personnel and of the need to demonstrate the organization of health services that are appropriate to different cultural patterns or structures, WHO is planning the establishment of regional training centres. In the All-India Institute of Hygiene in Calcutta, WHO and UNICEF are assisting in the development of the Maternal and Child Health, Health Education and Nutrition Departments and in strengthening the field practice services of the Institute, both urban and rural. While in the first phase the Institute will train largely Indian students it is hoped that it will soon be developed into a regional training centre. A similar development is envisaged for the Eastern Mediterranean and for the Latin American areas. An additional resource for training in social pediatrics and related subjects is the International Children's Centre in Paris, established by the French Government with the assistance of UNICEF.

In other instances totally different approaches to the problems of maternal and child health have been necessary. The following examples may be of particular interest.

A specialist in the production of whooping cough vaccine has visited the United Kingdom to advise on methods of production. She also visited Brazil, Chile and Colombia and gave assistance in the planning of immunization campaigns against whooping cough and diphtheria, which are now being carried out as joint WHO/UNICEF projects.

Another WHO consultant made an intensive study of the problem of infant diarrhoea in Finland, and as a result of his recommendations, the national administration, with the financial help of UNICEF, is undertaking a general programme of hygiene and sanitation measures, particularly a revision of the methods of milk pasteurization.

In France, following consultations with WHO experts, details of a national premature

baby programme which includes 25 centres, have been worked out, and will be implemented with the financial help of UNICEF. Similar premature baby projects are being carried out as UNICEF/WHO activities in Czechoslovakia, Poland, Finland, Yugoslavia and Italy.

Two teams, each consisting of three experts on the care of children with infantile paralysis, were recruited and sent out, one to Chile and one to India. In each case, WHO provided the team for three months and the country continued the service longer.

Another problem of childhood which is under specific attack is tuberculosis. WHO has, through its Expert Committee on Tuberculosis and the special panel on BCG, provided technical standards for the International BCG Campaign of the Joint Enterprise (UNICEF and the Scandinavian countries). Millions of children all over the world have been vaccinated by the teams of doctors and nurses provided by the Joint Enterprise, particularly in Europe, North Africa, and the Eastern Mediterranean countries. With the withdrawal of the Scandinavian partners in Joint Enterprise, WHO is now taking over the technical responsibility for new UNICEF/WHO BCG projects, and is carrying forward research on the efficacy of BCG vaccination at its Tuberculosis Research Office in Copenhagen. Until more is known as to the extent of immunity conferred by vaccination, WHO believes that BCG should not be depended on as the only measure used to control tuberculosis. In its present work with UNICEF, therefore, WHO is emphasizing the integration of BCG vaccination programmes into the over-all tuberculosis control programme.

In the field of Mental Health, WHO has helped in establishing a child guidance programme in the Philippine Islands. Special consultants, Dr. Bovet of Switzerland and Dr. Bowlby of the United Kingdom, have studied and prepared monographs on the psychiatric aspects of juvenile delinquency and separation of children from their parents. These monographs are a contribution to a study which the UN Division of Social Activities has been making on these two subjects, and are a basis for further co-operative programmes of action.

One of the most widespread nutritional disorders in tropical and sub-tropical areas is a syndrome, at present ill-defined but known by various names such as "kwashiorkor," malignant malnutrition, infantile pellagra, etc. The highest incidence occurs in infants and young children, and the mortality rates in some parts of Africa are disturbingly high. Two FAO/WHO consultants have studied this problem for some months in a wide range of African countries with a view to establishing its cause and ways of overcoming it in those countries where it is a serious problem.

For the too often neglected handicapped child, joint planning has gone ahead between WHO and the UN Division of Social Activities and the other specialized agencies concerned (ILO and UNESCO). A consultant jointly appointed by the UN and WHO has studied the problems of physically handicapped children in various European countries. Joint programmes of rehabilitation have been organized with UNICEF assistance in Austria, Bulgaria, Czechoslovakia, France, Greece, Italy, Poland and Yugoslavia. In addition a two months' course in rehabilitation of crippled children is being organized in the United Kingdom jointly with WHO, UNICEF, UN Division of Social Activities and ILO for further training of technical personnel connected with these projects.

In the Headquarters Office in Geneva, the Maternal and Child Health Section is concerned with the co-ordination of all field work being carried on in the regions, and takes

an active part in the recruitment of personnel to be employed on the international teams and of the short-term consultants who make special studies or surveys. The Section keeps in close contact with the Maternal and Child Health Regional Advisers, and is continually available to support them in their work and give them technical help as necessary.

A Clearing House Unit of Information first initiated in the Maternal and Child Health Section, and now broadened to cover the work of the Division of Organization of Public Health Services, will have as one of its functions to prepare documents and other materials on maternal and child health, or other fields, which can serve as a basis of information to the field advisers for adaptation by particular countries to their different patterns of culture and health service structure.

One of the important ways of establishing general guiding principles of work is through the convening of expert committees. In January 1949 the first WHO Expert Committee on Maternal and Child Health was convened in Geneva. This committee, composed of experts from nine different countries emphasized that a strong and balanced programme of maternal and child health can make essential and lasting contributions to any country or community. It made recommendations concerning the types of work for mothers and children which countries should develop and to which WHO could give assistance, ranging from the most fundamental and simple to the most far-reaching and complex. The experts emphasized the importance of legislation to establish maternal and child health units within or co-ordinated with national or local public health administrations, the necessity for improved training of health workers, and the need to stimulate research on various conditions affecting the health of children. Other Expert Committees and groups convened by WHO have made recommendations about more specialized aspects of maternal and child health—i.e., common communicable diseases of childhood, prematurity, and school health services. Furthermore, the Expert Committees on Mental Health, Nutrition, Nursing, Environmental Sanitation, Venereal Disease and Tuberculosis have dealt with the problems of children in these fields. In 1951 a Committee of Experts will meet to discuss Maternity Care. Joint Committees organized with the UN and the other specialized agencies concerned will discuss in 1951 the Crippled Child and in 1952, the Pre-School Child.

These, then, are the types of maternal and child health activities which WHO is carrying on. I have tried to indicate where they belong in the organizational framework of WHO, and to illustrate how they attempt to carry out the aims and policies of the World Health Organization.

There are, of course, many problems which confront WHO in connexion with these activities. Perhaps the most important is the question—what is being accomplished? WHO's plans are being laid on a long-term basis. Many of the projects in the field of maternal and child health have been underway less than a year. Only a few have been active as long as 1½ years. It is not possible to demonstrate dramatic results in this length of time, but it is hoped that sooner or later evidence will be available to show, among other things, a falling maternal and infant mortality rate in those areas where these rates are now high; a lowering in the incidence of prematurity and handicapping conditions where these are health problems of relative importance; and a statistical decrease in those diseases which are generally considered to be preventable. It is evident at the present time, however, that certain actions in the field of health must be under-

taken irrespective of our ability to demonstrate their results with statistical accuracy. Experience has taught us that favourable results, in the reduction of infant deaths, for example, may be expected if specified steps are taken. In the long run, however, methods of evaluation of success or failure must be instituted, and planning of selected types of projects must be such as to foresee such evaluation. In this planning process increased emphasis must be placed on ascertaining the economic, social and cultural factors that will make for success or that will act as barriers to the achievement of the child health goals that are sought. The work achieved so far has taught us a great deal about the importance of cultural patterns and the necessity of building health structures within these existing patterns and not superimposed on them.

While the economically underdeveloped areas face enormous handicaps, a number of them have high standards of culture and practically all of them have, from the point of view of child health, an enormous advantage in their almost untouched pattern of strong family life. If these countries, therefore, could achieve their over-all development within a structure that will not destroy the strength of the family and community pattern, they would have a much better chance of escaping the mental health problems with which highly developed societies are faced today.

It is to the attainment of these goals that WHO, within the framework of the UN, is devoting its efforts.

LOUIS VERHOESTRAETE, M.D.
Geneva, Switzerland

TRENDS

By JOHN P. HUBBARD, M.D., *Contributing Editor*

This and subsequent descriptions of events and trends are intended to be unbiased and factual, presenting both sides of controversial matters so that conclusions may be formed from critical appraisal of things as they are. These statements do not necessarily reflect the opinion of the writer nor are they to be interpreted as official opinion of the Academy.

THE CHILDREN'S BUREAU LOOKS AHEAD

By KATHERINE BAIN, M.D.

PERHAPS instead of saying "The Children's Bureau looks ahead," one might more correctly say that the Children's Bureau looks around at what is happening and peers ahead into the fog. For though one can identify many problems arising out of the emergency, their outline, bulk and duration are still indistinct. Many of the words we are using today are those we learned to use 10 years ago—day care, civil defense, evacuation. But the emergency of 1951 is far different from that of 1941. Then we were faced with a clearcut objective which could be mastered by rapid, all-out effort. We had before us something to be done quickly and finished. Today no such well-defined goal is visible. All one can say is that it looks like a long pull—not five years, but perhaps 15 or 20. In a quick dire emergency there is justification for abandoning some basic services. In view of the long pull ahead of us now, it would be suicidal to fail to meet basic needs of children. This country cannot afford to sacrifice one generation. In World War II our potential allies far outnumbered our enemies. Today our potential enemies far outnumber our friends. Each individual American is an asset which needs to be conserved.

Though the size of the emergency, its speed of build-up, and its ultimate duration are unpredictable at this moment, nevertheless we are *in* it, and planning has to take place. The Children's Bureau is faced with problems for which answers must be found. Emergency activities which bring back memories of 10 years ago are beginning to compete with basic programs for staff time. Day care of children of working mothers is again a public issue. Manpower policies, now being formulated, may greatly influence the pressures for employment of women. The need for day care, though high-lighted by an emergency, is part of a long-time trend in this country toward increased employment of married women. Whatever one's personal views may be about women with children working, the practice has become wide-spread and is now part of our culture. In the past, public programs of nursery education have been developed primarily as an accommodation to adults—in the 30's to make work for unemployed women, and in the 40's to get women into factories. Perhaps this time the needs of children will come first. In January 1951 the Children's Bureau and the Office of Education jointly called together a group of educators and social workers to discuss the present situation, to review past experience, and to recommend policies in the management of day-care programs which would insure their serving children's needs as well as those of adults. The Maybank bill, Senate bill

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349, to provide aid for community facilities and services (in some ways like the Lanham Act) has been introduced into the Congress. Under this bill, were it to pass, day care services could be provided. How great the need will be for these services still remains to be seen. The Children's Bureau, together with the Office of Education, is exploring needs and ways to meet them.

Civil defense is with us again, though in quite different terms from 1941. Its focus today is on emergency care in the event of bombing. Morale building through civilian participation in drives and in community activities is not part of the present civil defense program. The Children's Bureau has already helped the Federal Civil Defense Administration to prepare guide materials for the use of State and local civil defense offices in laying plans for the protection of mothers and children in the event of enemy attack. Evacuation of children from danger areas, plans for which were so well elaborated last time, is not a major element in the present planning. The experience of Britain in World War II and the type of bombing to which we may be subjected in the future have changed the thinking. Though civilians may have to be moved, wholesale anticipatory evacuation of children from our large cities is not contemplated.

A new term has been added to our vocabulary, "claimant agency." Under an Executive Order, certain agencies of the Federal Government have been designated to act as spokesmen for civilian needs. The Federal Security Agency under this order is claimant agency for health, welfare, recreation and education. The Public Health Service has assigned staff to determine the needs of hospitals and institutions for construction materials, equipment and medical supplies. Children's Bureau staff is working with the Public Health Service on the special maternity and pediatric needs.

Community "sore spots" are beginning to appear just as they did in 1940 and 1941. The Savannah River project is creating great problems of sanitation, health, housing and education over a wide area involving several States. The new Paducah area project will bring on similar problems. The field staff of the Children's Bureau has contributed time to surveys of these areas and to planning for meeting the special needs of children who have migrated there with their families.

What of EMIC? Inquiries are coming to the Bureau from doctors, from State health departments and from citizens about a revival of the Emergency Maternity and Infant Care program. Care of infants of service men seems not yet to be a great problem, but wives of service men are pregnant and doctors are asking who is to pay for their care. The Children's Bureau policy has been to keep its eyes open for evidence of need, to listen, and to be ready with the facts in case of need. At the present time (Feb. 15, 1951) no position for or against a new EMIC has been taken. The march of events will have to determine that position.

These and other emergency activities have been added to the work load of the Children's Bureau staff, a load already increased by more funds for grants to State with no more funds for administration. In 1950 the Congress authorized a higher ceiling for grants, though it appropriated less than the amount authorized.

Funds for Grants-in-aid to States

	1950	1951	1952 President's Budget (now before the Congress)
Maternal and Child Health Services	$11,000,000	$13,200,000	$13,120,500
Services for Crippled Children	7,500,000	9,975,000	11,927,700
Child Welfare Services	3,500,000	7,075,000	7,951,800

States are planning to use the increased funds in a variety of ways. Programs for children with epilepsy, with cerebral palsy, and with hearing defects are being started or expanded in many States. Many of the States have registered two primary needs for which some of the increased funds should go—training of personnel and evaluation of services. Shortage of staff is the single greatest obstruction to the "extension and improvement of services for mothers and children," the purpose for which the funds are granted. At a recent advisory committee on training, the consultants stressed the need for using increased amounts of MCH and CC funds for training. They placed the emphasis on training medical personnel, rather than on nursing, medical-social or nutrition, and especially on training in growth and development.

Second only to training is the increased interest in evaluating programs. Services are provided year after year without enough thought about changing emphases. Are the services being provided for children "good"? Do they accomplish the end for which they are designed? Perhaps no type of research is more difficult than program evaluation, because the criteria of success are hard to define and the tools for measuring success are poor indeed. In spite of difficulties we may anticipate that more States will be interested in setting up studies to try to determine adequacy and effectiveness of what they are doing.

At the close of the war it looked as though the Children's Bureau might push ahead on its own research program. Legislation was introduced into the Congress by Sen. Paul Douglas of Illinois for a program of grants to institutions and universities for research in child life. Though hearings were favorable, no action was taken. New bills along the same line have been introduced into this Congress by Mr. Douglas, Rep. Richard Bolling and Rep. Jacob K. Javits.

We came out of the last war a research-conscious people. One of the fundamental differences between a dictatorship and a democracy is in the attitude toward investigation. A dictatorship accepts the status quo as right, final, and not to be questioned. A democracy constantly questions, seeks new ways, hunts new knowledge. We learned the great value of basic and applied research in the physical and biologic sciences and to some extent in the social sciences. The layman knows now it is possible to study people and find out what makes them "tick." With a long pull of many years ahead of us, it is unthinkable that we abandon or even curtail our search for knowledge.

The White House Conference in December 1950 was one stage in a movement which began two years ago in grass-root planning and which will go on with increased momentum in State and local action. In terms of the development of important fact-finding material and mobilization of nationwide attention on children and youth, the Conference has already made an important contribution to the children's field. As in the instance of the Academy of Pediatrics Study of Child Health Services, it is not yet possible to measure adequately the full effects of this great enterprize. But if citizen interest is an indication of impending action, communities will see real change.

Everything we do in any agency today needs to be looked at critically to determine its value to society and its relative importance. Changes in program emphasis are inevitable. What we must not do is classify as "frills" or "luxury" the basic needs of children. Whether one argues from humanitarian motives, from sentiment, or from hardboiled common sense, we know children are not expendable. We cannot afford to waste them.

NEWS AND ANNOUNCEMENTS

The Michael Reese Hospital Postgraduate School will offer a one week course in "Recent Advances in Pediatrics—Diagnostic and Therapeutic Measures." This full time, intensive course will meet from May 21st to May 26th, 1951. Clinical and didactic material will be presented by members of the Department of Pediatrics and cooperating departments. For further information, address the Dean, 29th Street and Ellis Avenue, Chicago 16.

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A 6 months' fellowship is available at the Seizure Unit of the Neurological Institute, Children's Medical Center in Boston. This provides experience in electroencephalography and in the diagnosis and treatment of epilepsy of both office and clinic patients. For additional information, address Dr. William G. Lennox, 300 Longwood Avenue, Boston 15.

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Dr. Clifford G. Grulee, Executive Secretary of the American Academy of Pediatrics, has received the Annual Award made by *Parents Magazine* for outstanding service to children. The presentation was made in New York City on Jan. 31, 1951.

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Dr. Jacques M. Lewis of New York City, died on Jan. 29, 1951. He was a fellow of the American Academy of Pediatrics.

BOOK REVIEWS

MICROPHTHALMOS AND ANOPHTHALMOS WITH OR WITHOUT COINCIDENT OLIGOPHRENIA; A CLINICAL AND GENETIC STATISTICAL STUDY, Torsten Sjorgen and Tage Larsson, Acta Psychiatrica et Neurologica, Supplementum 56, 1949, 15 Swedish kroner or 21 Danish kroner (about $3.25), 103 pp.

Developmental defects resulting in microphthalmos or anophthalmos with a total incidence of 0.025 per 1000 are responsible for approximately 8% of the cases of blindness that reach school age in Sweden. This monograph is a detailed clinical and genetic study of 137 such individuals of which 58 had an associated mental deficiency. By what appear to be adequate approximations, it was estimated that the entire group of 137 cases represent from 80 to 90% of all such cases in Sweden as of 1947. Individual analyses are given of those with mental deficiency and those of normal intelligence. The degree of mental defect when present is in most instances profound. Associated with the mental deficiency are many other developmental anomalies sùch as cataracts (24%), coloboma (19%), epilepsy (28%), spastic diplegia (10%), skeletal defects, such as club foot, kyphosis, scoliosis, (70%). Microcephaly, congenital heart disease and deaf-mutism, however, were extremely rare or absent in this material. Toxoplasmosis, syphilis and maternal german measles seemed to be adequately excluded as etiologic factors. The statistical study indicated strongly that both groups, those with and those without mental deficiency, were primarily genetically determined, the former being most likely of a partially sex-linked mode of inheritance with low degree of expression. The latter occurred to a certain extent as a dominant Mendelian character, although it was not unlikely that exogenous factors may also have been involved in both groups. The study also indicated clearly that the group with mental deficiency probably represented a distinct homogeneous disease, either completely separate from the group without mental defect, or perhaps involving a question of linkage not at present understood.

Since the conditions described are extremely rare, the monograph will have limited appeal. The technic of the study, however, represents a model of genetic investigation.